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Environmental Protection Agency, Washington, DC

Sep 84

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The USEPA is charged, through a series of laws and executive orders, with reducing air and water pollution and finding a comprehensive approach to solving other environmental problems associated with toxic and hazardous substances. EPA's Office of Research and Development funds research, development and demonstration efforts in support of EPA's mandate. This is done through research grants, cooperative agreements, and negotiated contracts. Research grants are utilized mainly in support of investigator-initiated, long-range research activities which are subject to a highly competitive, extramural peer-review, and in-house priority process. The long-range agenda commenced in FY 1980 with a research grants activity. The contents of this Summary of Awards includes essentially all such grants issued since the program's inception as the Office of Research Grants and Centers, in the Office of Exploratory Research located in EPA's headquarters, Washington, DC.

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Office of Exploratory Research
Office of Research and Development
U.S. Environmental Protection Agency
Washington, DC 20460

1-a

Foreword

The Environmental Protection Agency (EPA) is charged, through a series of laws and executive orders, with reducing air and water pollution and finding a comprehensive approach to solving other environmental problems associated with toxic and hazardous substances. EPA's Office of Research and Development funds research, development, and demonstration efforts in support of EPA's mandate. This is done through research grants, cooperative agreements, and negotiated contracts. Research grants are utilized mainly in support of investigator-initiated, long-range research activities which are subject to a highly competitive, extramural peer-review, and in-house priority process. The long-range agenda commenced in FY 1980 with a research grants activity. The contents of this *Summary of Awards* includes essentially all such grants issued since the program's inception as the Office of Research Grants and Centers, in the Office of Exploratory Research located in EPA's headquarters, Washington, D.C.

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Title:

Degradation Kinetics of Nonpolar Organic Substrates by Microorganisms

Principal Investigator:
D. K. Button

Institution:
University of Alaska

Project Period:
2 years

Budget:
(2 years) $197,157

EPA Project Number:
R 808176

Summary:

Nonpolar substrates are known to accumulate in the aquatic environment. These hydrocarbon-rich compounds are metabolically oxidized in the water column by aerobic microorganisms to give mostly CO_2 and cell material. The enzymology of metabolism is well described. However, views differ as to whether the compounds are actively transported into the organism, simply diffused into the cytoplasm, or are largely extracted from oil phase droplets by the cells. All but a few studies of hydrocarbon metabolism rates include an oil phase so that the concentration dependency of growth rate on dissolved hydrocarbons is without a convincing description. With the emergence of hydrocarbon solubility data and related theory, it has become feasible to consider the kinetics of dissolved hydrocarbon metabolism by microorganisms. Measured marine hydrocarbon concentrations are well below saturation concentrations. Suspended sediment has little sorptive capacity for hydrocarbons and little effect on their rates of metabolism, so that the principal metabolic route is directly through the dissolved phase. Since procedures for measuring microbial metabolism kinetics of nonpolar substrates at low concentrations have been developed, various patterns in the associated rate constants (as affected by structural characteristics of the contaminants, the influence of additional similar contaminants, the presence of dissimilar organic compounds, the nature of microflora, and temperature) will be evaluated. The resulting kinetic constants will be useful in predicting the stability of many important contaminants in the aquatic environment.

Title:

Biological Indicators of Oil Pollution for Alaskan Marine Environments

Principal Investigator:
David G. Shaw

Institution:
University of Alaska

Project Period:
2 years

Budget:
(2 years) $157,795

EPA Project Number:
R 807918

Summary:

The objective of this project is to evaluate a selected group of potential biological indicators of oil pollution for use in Alaskan and other cold climate marine environments. Calculations will be made regarding the energy charge, the scope for growth, and the shell growth in bivalve mollusks using controlled laboratory tests to show the relationship of the indicator parameters to known oil exposure. These tests will then be followed by *in situ* field tests of the indicators in areas (Port Valdez and Cook Inlet, Alaska) known to receive chronic inputs of oil. The species used in this work will be *Macoma balthica* (a deposit feeding clam) and *Mytilus edulis* (the common mussel).

Title:

Development of In Vitro *Bioassays for the Detection of the Effects of Environmental Toxicants on Immunity in Humans*

Principal Investigator:
Floyd D. Wilson

Institution:
Battelle Memorial
Institute
Columbus
Laboratories

Project Period:
2 years

Budget:
(2 years) $163,920

EPA Project Number:
R 808362

Summary:

This project proposes to develop and perfect a series of short-term *in vitro* bioassays for the quantitation of toxic effects on immunity in humans. Evidence of the effects of a wide variety of chemical and physical agents suggests that the environment in association with hazardous wastes from energy production, industrial processes and other mechanisms have the potential of toxic effects on the immune system. Determination of these effects will be made through an assay development system which will be structured in part on existing methods that allow for the cloning of lymphocytes in semisolid cultures. The research will include comparisons on the sensitivity of the new methods to more standard techniques in their ability to measure the effects produced by a routine battery of environmentally important physical and chemical agents with demonstrated immunotoxic properties.

Title:
Waterborne Rotavirus Infections

Principal Investigator:
David Y. Graham

Institution:
Baylor College of Medicine

Project Period:
2 years

Budget:
(2 years) $168,008

EPA Project Number:
R 808333

Summary:

Water quality is of increasing public health concern and the detection of new contaminants in water supplies mandates investigation of their public health significance so that specific guidelines for maximum allowable levels of contamination can be formulated. The overall objective of this proposal is to determine the minimal infectious dose of the important gastroenteritis virus, rotavirus. This study aims (1) to utilize a sensitive *in vivo* biologic assay method to study low dose rotavirus infections using miniature swine piglets and porcine rotavirus; (2) to identify the minimal infectious dose of virus ingested in water; and (3) to assess the effect of viral dosage on the pathophysiology of the disease. These studies will assist public health officials to determine future recommendations for standards on virus contamination and expand existing data on the enteroviruses, particularly the rotavirus.

Title:
Evaluation of Electrode Systems for Chlorine Determination in Wastewater Matrices

Principal Investigator:
James T. Kinard

Institution:
Benedict College

Project Period:
2 years

Budget:
(2 years) $89,589

EPA Project Number:
R 807425

Summary:

This research is designed to determine the response time, run-to-run reproducibility and overall precision, and accuracy of commercial chlorine electrode systems in wastewater matrices. The scope of the proposed work will also encompass the establishment of operating limitations of these systems and the development of an electrode method which would be capable of distinguishing total, free, mono-, di-, and other combined chlorine species in aqueous media. The approach will involve the determination of data for each system in a distilled water matrix and various wastewaters before and following the application of various forms of chlorine at different pH levels. Subsequent results with the amperometric titration method will be compared.

Title:

Kinetics and Mechanisms of SO_2 Oxidation in Bulk Aqueous Solution and Submicron Aerosols

Principal Investigator:
Michael R. Hoffmann

Institution:
California Institute of Technology

Project Period:
2 years

Budget:
(2 years) $244,047

EPA Project Number:
R 808086

Summary:

An experimental and theoretical study of SO_2 oxidation in aqueous solutions is proposed. The kinetics and mechanisms of SO_2 oxidation in bulk aqueous solutions will be studied. Catalytic autoxidation of SO_2 in the presence of trace metal and organometallic complexes; the concomitant production of H_2O_2, $HO_2\cdot$ and $HO\cdot$; the catalytic oxidation of SO_2 by H_2O_2 in the presence of trace metals (i.e., Fenton's Reagent); and the noncatalytic oxidation of SO_2 by H_2O_2 at $pH < 4.5$ will be studied. Experimental techniques will include Stopped-Flow Spectrophotometry, Ion Chromatography, Temperature Jump/Stopped Flow Relaxation Kinetics and Specific Ion Potentiometry. Electron Spin Resonance Spin Trapping, Nuclear Magnetic Resonance and Mossbauer Spectrometry will be used to determine the relative contribution of radical, enzyme-like, surface and polar reaction pathways to the overall liquid phase oxidation rate of SO_2. In a complementary study, other researchers at Caltech will measure growth rates of monodisperse aerosol droplets by liquid phase SO_2 oxidation in a CFSTR reactor. Gaseous concentration, dew point, and initial droplet composition (pH, solute) and size are important variables to be investigated. A final objective of this study will be an attempt to predict measured droplet growth rates from rate constants and rate laws inferred from the solution phase studies. This work will have application in understanding gas-to-particle conversion of SO_2 in polluted air, as well as in removal of SO_2 from stack gas scrubbers.

Title:
Respiratory Function of Five Species of Mammals with Air Pollutant Injury

Principal Investigators:
Jerry R. Gillespie,
Allen J. Marr

Institution:
University of California

Project Period:
2 years

Budget:
(2 years) $582,013

EPA Project Number:
R 807661

Summary:

The objectives of this project are (1) to evaluate the comparative respiratory physiology of the laboratory animals frequently used to study the health effects of air pollutants; (2) to study the comparative respiratory function response of five species of laboratory animals to a well characterized environmental air pollutant, ozone; (3) to investigate a new non-evasive pulmonary function technique to be used to detect early airway injury caused by air pollutants; (4) to expand the scope of dynamic pulmonary function test available to study airway function in small laboratory animals; and (5) to develop quantitative and qualitative respiratory function and pathophysiological data in animals which will enable correlation prediction parameters in human beings.

Title:

Interaction of Carbon Monoxide with Noise and Heat Stress: Psychological and Physiological Changes

Principal Investigator:
Jeffrey A. Gliner

Institution:
University of California

Project Period:
2 years

Budget:
(2 years) $253,572

EPA Project Number:
R 808177

Summary:

The object of this proposal is to examine the effects of carbon monoxide alone and in combination with noise stress and heat stress on man's ability to process information from the environment (i.e., ability to perform psychological and normal work tasks). The ability to process information under these conditions is considered critical for industrial and environmental safety. The approach undertaken is in the context that man actively processes information from the environment and then acts on that information by making a decision. Skilled movement is then performed through the effector system to meet the environmental demands. Since man has only a limited attention capacity, it can be determined that in the processing system a breakdown will occur by utilizing psychomotor tasks which divide the subject's attention and hence overload either the perceptual or motor system. Two different types of tasks will be utilized. One task will be a tracking task performed concurrently with a peripheral vision task. The emphasis will be on perceptual processes. The other divided attention task will be a reciprocal tapping task in combination with a digit subtraction task. The emphasis of this task is on the effector system. Using these two types of tasks will determine whether carbon monoxide alone or in combination with heat and noise stress affects man's ability to process information. It will also establish where in the system these stresses cause a deterioration in performance.

Title:
Particle Size Distribution Dynamics During Coagulation

Principal Investigator:
James R. Hunt

Institution:
University of California

Project Period:
2 years

Budget:
(2 years) $109,816

EPA Project Number:
R 808382

Summary:

The main objective of this project is to test the validity of theoretical particle size distribution predictions for coagulating particles under conditions typically encountered in water and wastewater treatment. Previous theories for particle coagulation kinetics were limited to the initial coagulation kinetics of particle suspensions uniform in size. More recently, theoretical predictions for coagulation kinetics in continuous particle size distributions were obtained, and experiments conducted with clay particles in seawater verified the Brownian and shear coagulation predictions. This project proposes to accomplish three tasks: (1) the development of a technique for measuring aggregate particle size distributions, which is complicated by the weak aggregate formed during coagulation. Aggregate size distributions will be measured using the electrical resistive pulse sensing technique with a microprocessor controlled data acquisition system; (2) to design and conduct experiments for testing the validity of the particle size distribution predictions. Well characterized particles destabilized by conventional coagulanats will be coagulated under uniform fluid shear, and the particle size distribution will be measured over time; and (3) to examine, theoretically and experimentally, the effect of high fluid shear rates on particle aggregation and breakup.

Title:
The Entrainment and Deposition of Sediment in the Great Lakes

Principal Investigator:
Wilbert Lick

Institution:
University of California

Project Period:
2 years

Budget:
(1 year) $103,271

EPA Project Number:
R 807742

Summary:

The two main purposes of the research are: (1) to quantitatively determine the parameters on which entrainment and deposition depend, and (2) to determine the entrainment and deposition rates for a range of sediments representative of those in the Great Lakes. Laboratory experiments to determine the entrainment and deposition rates of fine-grained sediments from Lake Erie are being performed. Results are being compared with field data from the Western Basin of Lake Erie. Parameters on which entrainment and deposition depend (bottom stress, compaction, mineralogy, bioturbation, manner of deposition) are being investigated.

Title:

Environmental Chamber Studies of NO_2, NO_3, HONO, N_2O_5, and HNO_3 by in situ *FT-IR and Differential UV-Visible Spectroscopy*

Principal Investigator:
J. N. Pitts, Jr.

Institution:
University of California

Project Period:
2 years

Budget:
(1 year) $159,478

EPA Project Number:
R 807739

Summary:

A three-year research program using state-of-the-art spectroscopic and environmental chamber techniques to investigate the chemistry of NO_x species with emphasis on NO_3, N_2O_5 and HONO has been proposed. This research will assist in the development, validation, and application of photochemical smog models for more effective air pollution control. The experiments in this research include the use of (1) a 5800-liter thermostatted, evacuable environmental chamber facility; (2) a Fourier transform infrared (FT-IR) spectrometer which is interfaced to an *in situ* white cell in the chamber; and (3) the newly developed Differential Ultraviolet-Visible Spectrometer (DUVVS) system constructed at Statewide Air Pollution Research Center (SAPRC). Emphasis will be placed on adaptation of a multipass DUVVS system for operation in the environmental chamber in a crossed-beam configuration with the FT-IR system. This will permit simultaneous *in situ* measurements of NO_2, NO_3, N_2O_5, HONO, HNO_3, O_3 and other pollutants. Through fundamental spectroscopic and kinetic studies involving NO_3 and N_2O_5, the following reactions will be observed: (1) the absolute extinction coefficient for NO_3; (2) the direct measurement of $NO_2 + NO_3 \rightleftharpoons N_2O_5$ equilibrium constant as a function of temperature; (3) the N_2O_5-O_3-NO_x system; and (4) measurement of rate constants for reactions of NO_3 with CO and selected organics as a function of temperature. Concurrently, heterogeneous processes occurring in the Teflon-lined chamber, including the "unknown chamber radical source" rates of heterogeneous hydrolysis of N_2O_5 and heterogeneous production of HONO will be studied.

Title:
Performance of Overland Flow Wastewater Treatment Systems

Principal Investigator:
Edward D. Schroeder

Institution:
University of California

Project Period:
2 years

Budget:
(2 years) $110,899

EPA Project Number:
R 807757

Summary:

The objectives of the proposed research are: (1) to determine the effects of precipitation on overland flow process performance; (2) to determine the final fate of suspended solids removed in the overland flow process; and (3) to investigate nitrogen removal control techniques and develop operating procedures to optimize nitrogen removal by overland flow. Pilot and full-scale research studies will be conducted. The pilot studies will be performed at the Pilot Overland Flow System, University of California (Davis). Results from these studies on precipitation effects and nitrogen removal will be verified at the full-scale overland flow system, the City of Davis Wastewater Treatment Plant. Precipitation pilot studies will involve overhead application of dionized water to simulate rainwater. Precipitation field studies will incorporate control plots to determine runoff water quality from slopes that have not received wastewater. Studies will involve measuring at frequent intervals (1) the mass and (2) the organic content of suspended solids deposited down the slope and those found on the overland flow slope. Nitrogen studies will involve (1) investigation of flooding the lower end of the slopes to promote denitrification and (2) investigation of various application and drying periods to determine if nitrogen removal can be maximized through operating procedures.

Title:
Fuel Sulfur-Fuel Nitrogen Interactions in Flames

Principal Investigator:
Owen I. Smith

Institution:
University of California

Project Period:
1 year

Budget:
(1 year) $99,754

EPA Project Number:
R 808352

Summary:

The objectives of this investigation are to identify the mechanism by which the conversion of fuel-N to NO is enhanced by the presence of fuel-S compounds in rich flames and to collect kinetic data on the most important reactions in the mechanism. To do this, SO_2 will be added to rich hydrocarbon/oxygen/argon flames doped with cyanogen. A laminar, premixed flame will be utilized at pressures of ≤ 1 atmosphere. The flame will be sampled as a function of distance from the burner face by means of nozzle-beam-skimmer system. The sample analysis will be done by mass spectrometry. Species profiles for flames with or without the SO_2 addition will be compared in an effort to identify the mechanism of interaction. Currently, equipment is being fabricated. The collection of species profiles is expected to begin in May of 1981, with attention initially focused on known intermediates in the fuel-N to NO conversion mechanism, particularly the NH_i species ($i=1,2,3$).

Title:

Probabilistic Analysis of the Escape and Environmental Establishment of Laboratory Organisms

Principal Investigator:
David R. Lincoln

Institution:
Carnegie-Mellon University

Project Period:
2 years

Budget:
(2 years) $126,909

EPA Project Number:
R 808317

Summary:

The discovery of the technique of recombinant DNA in 1973 created the opportunity for engineered living systems. The potential impact of these new genomes in the existing biosphere has not been well characterized. The assessment issues for examining the potential impact are complex, including the potential for environmental release during manipulation, establishment in favorable ecological niches, infection of existing life forms, and the release of harmful agents. It is proposed that a probabilistic framework based on fault tree analysis to examine the potential for these organisms to be released in laboratory or pilot plant scale conditions be developed. Within this framework, the interacting effects of organism quantities being handled, protocols being followed, the quality of technicians and the equipment designed to contain releases will be considered. Limited aspects of the environmental establishment of the organisms condition on their escape will also be examined. The probabilistic model will be coded for computer operation for ease of computation and evaluation. Major issues to be developed quantitatively include: (1) cumulative probability functions of releases to different environmental media (air, water, and solids) in different operating scenarios; (2) key functions and activities which have significant influence on escape and establishment probabilities; (3) sensitivity analysis to examine the effects of uncertainty and to identify future areas for investigation; and (4) cost-effectiveness analysis of major procedural or equipment changes. Results of the proposed systems approach are expected to bring new insights and understanding to these aspects of risk evaluation in applied genetics.

Title:

Efficacy of Hydrilla Control and a Stocking Model for Hybrid Grass Carp in Freshwater Lakes

Principal Investigator:
John A. Osborne

Institution:
University of
Central Florida

Project Period:
2 years

Budget:
(2 years) $112,532

EPA Project Number:
R 807961

Summary:

The primary objective of the research project is to determine the efficacy of the hybrid grass carp (cross between *Aristichthys nobilis* and *Ctenopharynodon idella)* in controlling submersed aquatic vegetation, principally *Hydrilla (Hydrilla verticillata)* and to use vegetation biomass data and stocking rates to determine the stocking rate model for the hybrid grass carp. Eight Florida lakes are being used in the study, ranging in size from 1 to 140 acres and all have *Hydrilla*. Baseline vegetation biomass at the peak of the growing season (October-November) was achieved at the onset of the project, and stocking rates will be constant for all lakes (per surface area). Seven of the lakes will be stocked with hybrid grass carp in Spring 1981, by the Florida Game and Fresh Water Fish Commission, while the eighth lake has been previously stocked by UCF. Growth trends for vegetation species, primary production data per species, and vegetation maps will be used to analyze the effect of the hybrid grass carp over a two-year period. If the hybrid grass carp proves to be an effective biological control agent for problematic submersed vegetation, the stocking rate model will allow for operational use of the fish by providing the basis for stocking to obtain control, while minimizing the number of fish and thus the cost of the operation.

Title:
Cell Death and Somatic Mutation in Teratogenesis

Principal Investigator:
Jeanne M. Manson

Institution:
University of Cincinnati

Project Period:
2 years

Budget:
(2 years) $315,123

EPA Project Number:
R 807774

Summary:

The objective of the research is to determine the outcome of DNA damage in embryonic cells exposed to a mutagenic teratogen during the organogenesis period. Tests will be conducted to examine whether DNA damage in embryonic tissues during this critical period leads to cell death of affected cells and/or heritable alterations (somatic mutations) in cells surviving teratogenic insult. Methods of measuring DNA damage and mutation in an isolated organ system of the mouse embryo, i.e., the limb buds, during the organogenesis period will be perfected. The persistence of DNA damage and mutation after *in vivo* and *in vitro* treatment with the mutagenic teratogen MNNG (methyl-N'-nitrosoguanidine) is being studied. The level and persistence of cell death in limb buds after treatment with MNNG is being measured and correlated with the extent of DNA damage and orientation. The DNA repair capacity of embryonic limb buds after teratogenic exposure to MNNG is being ascertained and correlated with the target-organ specificity of MNNG teratogenesis. Three methods will be employed to measure the effects in day 11 mouse embryo limb buds at different time intervals after exposure to MNNG: (1) alkaline elution analysis to detect alkali-labile sites in single-stranded DNA; (2) mutation selection tests with onabain and 6-thioguanine; and (3) HPLC analysis of limb bud DNA for methylated bases. It has been found that the sensitivity to MNNG-induced limb effects occurs in the following order: left hindlimb, left forelimb, right hindlimb, and right forelimb. Distribution studies using radiolabeled MNNG *in vivo* and organ cultures *in vitro* will be carried out to determine if the differential response is due to differences in delivery of the chemical *in vivo* or to the differences in susceptibility of the limb.

Title:

Effect of Turbidity and Other Factors on the Virus Disinfection Process Using Free and Combined Chlorine Forms

Principal Investigator:
Pasquale V. Scarpino

Institution:
University of Cincinnati

Project Period:
2 years

Budget:
(2 years) $173,500

EPA Project Number:
R 808248

Summary:

This research study is a direct extension of continuing efforts to learn more about factors influencing viral and bacterial water disinfection processes in the presence of particulate matter (i.e., turbidity) with forms of free chlorine and combined chlorine as the disinfectants. Turbidity in water will be created using (a) inorganic suspended solids as bentonite- and kaolin-type clay suspensions, and by hydrated aluminum oxide flocs formed by the coagulant aluminum sulfate $Al_2(SO_4)_3$ used in water and wastewater treatment; (b) organic suspended solids as Hep-2, BGM, and MA-104 tissue culture cells to simulate human intestinal tract cells that may be found in water harboring enteric viruses; and (c) human and/or animal fecal matter. The disinfecting capabilities of the chlorine forms will be evaluated using enteric viruses (e.g., poliovirus 1 and coxsackievirus A9) and animal rotaviruses (e.g., simian isolate SA-11) at different chlorine levels, temperatures, contact times, pH values, concentrations and types of particulate matter, ionic environments, and types of virus inocula (i.e., singles or aggregated). For comparative purposes, reference bacteria (i.e., both stock and naturally-occurring total coliforms found in sewage) and bacteriophages (e.g., f2, MS2, and T2) will be used. The effect of the disinfectants on enterovirus- and rotavirus-associated tissue culture cells, which simulate naturally found cell-associated viruses excreted from the intestinal tract of man, will be also evaluated. The studies of breakpoint chlorination will be extended to ascertain the effects of microbial kill of changing chlorine to ammonia and ammonia to chlorine ratios at varying pH (between 4.5 and 10) values and mixing rates during the process of formation of monochloramine and dichloramine, respectively. The chloramine disinfectant studies will be performed using the apparatuses of Scarpino and Sharp.

Title:
Physiological Aspects of Heavy Metal Uptake in Algae

Principal Investigators:
Joseph W. Rachlin
Thomas E. Jensen

Institution:
CUNY

Project Period:
2 years

Budget:
(2 years) $110,270

EPA Project Number:
R 807035

Summary:

Recognition of the risk of heavy metal contamination to aquatic life and its potential movement through the food chain have led to EPA requirements for monitoring of heavy metals at all sewage sludge dump sites. In order to better assess the fate of heavy metals in aquatic environments, both laboratory and field tests are proposed to examine algal uptake of heavy metals. Three basic tests will be performed in three separate systems. The three tests are: (1) determination of the presence of copper, lead and cadmium in algae by energy dispersive X-ray analysis; (2) qualitation and quantification by energy dispersive X-ray analysis; (3) quantitative analysis by either anoidic stripping voltammetry or atomic absorption spectrometry. The three systems are: (1) culture of algae native to the Hudson Estuary System and environs in the presence of selected heavy metals; (2) culture of similar algae in an "overplus environment" known to cause the formation of phosphate storage morphologues referred to as polyphosphate bodies; and (3) collection of algae from known metal contaminated sites. Benefits noted include a better understanding of the impact of heavy metals on the food chain via algal accumulation, and the relationship of phosphorus to algal incorproation of heavy metals.

Title:

The Sensitivity of Critical Life Stages of Benthic Meiofauna to Drilling Fluids

Principal Investigator:
John J. Lee

Institution:
City College of
CUNY

Project Period:
1 year

Budget:
(1 year) $36,287

EPA Project Number:
R 807214

Summary:

Meiofauna (nematodes, copepods, foraminifera, and similar sized [0.05-0.5mm] small organisms) are the most numerous of marine animals and play a key role in benthic food webs. These organisms consume and regulate benthic microfloral assemblages and serve as food for larger animals. Current research indicates that the time scales for the recovery of acutely stressed near-shore marine environments vary over wide ranges. Planktonic communities may recover depending upon prevailing surface conditions in relatively short periods (days, months). The longest impact, however, is on sheltered littoral zones and sublittoral benthos with only low rates of circulation. Meiofauna are among the most sensitive indicators of environmental quality because they live in direct contact and feed in benthic sediments. These organisms offer great potential for monitoring environmental impact on vulnerable and vital coastal ecosystem processes. Also, little is known of the sensitivities of critical life stages of meiofauna to the drilling fluids used in petroleum exploration. It is proposed through laboratory testing that data on the sensitivity of critical life of meiofauna will yield information relative to probable areas of impact, duration of impact, and recovery potential from the effects of drilling fluids. Their small size and easy cultivation in ordinary glassware, test tubes and flasks under common laboratory conditions (25°C) does much to recommend them as a potential bioassay tool. Five (5) species of benthic meiofauna will be used: (1) *Allogromia laticollaris* (Foraminifera); (2) *Nitocra typica* (harpacticoid copepod); (3) *Chromadorina germanica* (algal-feeding nematode); (4) *Rhabditis marina* (bactivorous nematode); and (5) *Euplotes vannus* (ciliated protozoan). These species were chosen because of their wide distribution, easy culturation, and minimal manipulation requirements. Procedures for developing the potential bioassay generally will follow *Standard Methods for the Examination of Water and Wastewater*. The LC_{50} and EC_{50} will be determined.

Title:
Fate in Established Ecosystems of Microbial Species Representing Potentially New Genomes

Principal Investigator:
Martin Alexander

Institution:
Cornell University

Project Period:
2 years

Budget:
(2 years) $134,918

EPA Project Number:
R 807688

Summary:

The objectives of this investigation are: (1) to determine the likelihood of persistence, survival and growth of microbial species (representing potentially new genomes) in model sewage, soil and fresh and marine water ecosystems; (2) to establish the environmental factors governing their persistence, survival or growth; and (3) to establish the reasons for decline or persistence. Taxonomically diverse microorganisms, both procaryotic and eucaryotic, have been selected for use. The recovery and enumeration of the test organisms will be achieved through the use of antibiotic-resistant mutants obtained from the parent test strains and selected media. (These have been developed for all the organisms to be studied.) Known numbers of the test organisms will be added to the model ecosystems, and then these ecosystems will be monitored by plate counts to assess growth, persistence or decline. The significance of biotic factors (i.e. predation, parasitism and carbon sources) and abiotic factors (i.e. temperature, pH, light and moisture) will be considered to explain population changes. Currently, the survival of *Agrobacterium tumafaciens* and *Staphylococcus aureus* when inoculated in soil is being investigated. Determinations as to persistence or lack of survival will be conducted through studies designed to ascertain if abiotic and biotic factors are responsible for the die-off of the organisms. Characterizations of factors governing the persistence of organisms that survive or grow will also be assessed.

Title:

The Removal of Heavy Metals by Activated Carbon Process from Water and Wastewater

Principal Investigator:
C. P. Huang

Institution:
University of Delaware

Project Period:
2 years

Budget:
(2 years) $125,189

EPA Project Number:
R 808141

Summary:

The major objective of this proposed research project is to develop an activated carbon process for the removal of heavy metals from water and wastewater. Laboratory experiments will be conducted using a batch reactor to study the mode and rate of adsorption and reduction between heavy metals (copper, lead, zinc, mercury, cadmium, nickel, cobalt and silver) and a nonmetal (arsenic and commercial activated carbons). The effect of complex formation on the chemical reactions will also be studied. Laboratory experiments involving chemical and thermal regeneration techniques will be conducted to regenerate the heavy metal-laden activated carbon. Strong acids, strong bases, wet thermal and combined chemical-dry thermal regeneration methods will be thoroughly studied.

Title:
A Single Gene Mutagenesis System in Drosophila

Principal Investigator:
P. Dennis Smith

Institution:
Emory University

Project Period:
2 years

Budget:
(2 years) $133,425

EPA Project Number:
R 807538

Summary:

Experiments are outlined for the development of a single gene mutagenesis system in *Drosophila* as a model for mutation induction by chemical agents in animal cells. The system to be examined is the *rosy* (*ry*) eye color gene, a genetic unit specifying the enzyme xanthine dehydrogenase (XDH). Extensive genetic and biochemical studies make this gene the most defined locus in *Drosophila*. It is an excellent candidate for studies of mutation induction which probe both genetic and biochemical events. Mutation studies will employ a group of chemical mutagens which includes methyl methanesulfonate, ethyl methanesulfonate, ethyl nitrosourea, nitrogen mustard, N-2-fluorenylacetamide and hydroxylamine. These agents have been chosen as representative of a defined series which vary systematically with respect to their modes of action, reaction mechanisms, types of lesions and presumed involvement with different repair mechanisms. Employment of these varied agents is designed to demonstrate the general utility of the *rosy* system for the detection of chemical mutagens. Forward mutation studies will lead to the production of *ry* mutant alleles from the enzymatically and electrophoretically defined ry^{+11} wild type allele. The frequency of mutation induction will be determined for each mutagen and newly-induced *ry* mutants will be subjected to genetic and biochemical tests. These tests will include examination of complementation behavior and determinations of level of XDH activity and production of XDH cross-reacting material. Reverse mutation studies will take advantage of an effective chemical selection procedure which will kill developing *rosy* mutants and allow specific isolation of ry^{+} revertants. Employment of the EMS-induced electrophoretically-defined ry^{601} mutant allele for reversion experiments will permit confirmation of revertants by both enzymatic and electrophoretic means. Additional experiments which couple the forward and reverse systems can be undertaken to examine the nature of the mutation events induced by specific chemical mutagens. The role of DNA repair processes in the production of mutational events by specific chemical mutagens will be examined. The introduction of excision-defective and postreplication repair-defective mutants into the *rosy* system will be designed to identify genes which control error-free or error-prone repair. These studies will be useful not only in defining fundamental mechanisms of mutation induction in higher eukaryotic cells but also in providing sensitive and well-defined tester strains for the identification of potentially mutagenic chemical agents. The employment of sensitive and biochemically defined tester strains in a multicellular, differentiated eukaryote which has the capacity for *in vivo* metabolic activation of procarcinogenic compounds offers a significant improvement in our ability to assess the risk of exposure of human populations to environmental chemicals.

Title:
Effect of Hazardous Waste on Human Nerve Conductivity, Growth and Maturation

Principal Investigator:
Joseph H. Highland

Institution:
Environmental
Defense Fund

Project Period:
1 year

Budget:
(1 year) $39,203

EPA Project Number:
R 807772

Summary:

The ultimate objective of the project is to determine whether the growth of children and peripheral neuropathy are reliable and sensitive measurements of chemical toxicity in a population living near a hazardous waste site. The residents of Love Canal, Niagara Falls, New York, will provide a test population since they have already demonstrated toxicity due to exposure to hazardous wastes and have a large enough population for statistical purposes. They will provide an extensive data base on occupational exposures, chronic illnesses, reproductive histories and other pertinent factors which will be useful for comparisons with the studies described below. A control neighborhood, near Love Canal, will be identified which has much in common with the Canal, but has no hazardous waste site. Growth and maturation in children and peripheral neurotoxicity will be studied and compared in the test and control populations. The practicality of such a test will be evaluated. A standard protocol will be devised and a study of a second population living near a hazardous waste dump will provide a validation study. Two potential sites are Gray, Maine, and Memphis, Tennessee, which differ from Love Canal ethnically, culturally and geographically.

Title:

Immunologic Studies in Asbestos Exposure

Principal Investigator:
Elliott Kagan

Institution:
Georgetown University

Project Period:
2 years

Budget:
(2 years) $455,483

EPA Project Number:
R 808252

Summary:

The objectives of this study are as follows: (1) to determine the contribution of immunologic processes to the pathogenesis of asbestosis; (2) to ascertain the effect of chronic asbestos inhalation on the functioning of the immune apparatus; and (3) to determine the morphologic and ultrastructural effects produced by asbestos inhalation in the lungs. The role of the pulmonary alveolar macrophage is considered crucial to the evolution of asbestosis. Attention will be given to the isolation, purification and characterization of a putative asbestos-related antigen on the surface of alveolar macrophages from unexposed control rats. Mechanisms of immune derangement occurring as a consequence of the inhalation of asbestos will be studied in a rat model. Conventional light microscopy and transmission electron microscopy will be used to study the effects of asbestos, another group to chrysotile asbestos, and another group will serve as age- and sex-matched unexposed controls. The exposures will last for 3 months. Alveolar macrophage membrane fractions will be evaluated for antigenic activity. Animal sera will be assayed for antibody activity against a putative asbestos-related antigen. Spleen lymphocytes will be used to test for evidence of cell-mediated immunity against an asbestos-related antigen. The functional integrity of the immune apparatus will be evaluated in all three animal groups using a series of immunologic procedures. Spleen lymphocytes will be used to monitor cellular immunity and serum will be used to assess humoral immunity. Animal exposures are to commence around mid-February, 1981.

Title:

Chlorination and Ozonolysis of Purine and Pyrimidine Bases in Water: Products, Pathways and Potential Health Effects

Principal Investigator:
J. P. Gould

Institution:
Georgia Institute of Technology

Project Period:
2 years

Budget:
(2 years) $99,951

EPA Project Number:
R 807367

Summary:

The objective of this project is to clarify the health effect implications of chlorination in water and wastewater treatment and to compare this process with its most commonly used alternative, ozonation. A group of related, biologically significant organic compounds, all purine and pyrimidine bases, will be treated with chlorine or ozone under conditions common to water and wastewater treatment. The resulting reaction mixtures will be chemically analyzed to identify and quantify specific reaction products. Reaction mixtures and pure products will then be biologically tested for mutagenic and carcinogenic activity using three test systems (the Ames test, tissue culture test and whole animal test). The following information will be gained from these studies: (1) exact pathways of reaction between the oxidants and the purine and pyrimidine bases will be clarified; (2) mutagenic and carcinogenic potentials of chlorination, as opposed to ozonation, will be assessed; (3) data from three different biological tests will be available for comparison; (4) knowledge of which compounds might be mutagenic or carcinogenic will be established; and (5) indirect data relative to the process of disinfection will be provided.

Title:

Electrochemical Detectors for High Performance Liquid Chromatographic Determination of Ultratrace Pesticide and Coal Phenolic Residues in Water

Principal Investigator:
James L. Anderson

Institution:
University of Georgia

Project Period:
2 years

Budget:
(2 years) $64,915

EPA Project Number:
R 808084

Summary:

The purpose of this project is to improve the analytical methodology for the determination of ultratrace pesticide and coal phenolic residues entering the water environment. Analyses will be based on high performance liquid chromatography (HPLC) with electrochemical detection. The pesticides to be studied (carbamates and ureas) will be chosen on the basis of projected future environmental significance as well as current limitations in analysis based on other techniques. A series of important pesticides will be screened by cyclic voltametric techniques to determine the general applicability of electrochemical detection in pesticide analysis. Initially, selected compounds which exhibit electroactivity will be analyzed as pure reagent solutions. These compounds will then be analyzed as authentic analytical standards, added to and recovered from various natural water samples at ultratrace levels. Finally, the compounds will be analyzed as environmentally occurring traces in natural water samples. Performance of the electrochemical detector will be compared with performance of a conventional ultraviolet detector. Optimized methods and instrumentation will be tested under routine use conditions to ascertain the potential for application by field workers to the determination of environmental pollutants affecting water quality.

Title:
Pulse-Jet Fabric Filter Performance

Principal Investigator:
David Leith

Institution:
Harvard School
of Public Health

Project Period:
2 years

Budget:
(2 years) $266,146

EPA Project Number:
R 808001

Summary:

Fabric filters are the most efficient devices available for the practical collection of many industrial dusts. Pulse-jet cleaned filters comprise about 40% of the filters sold. Presently, it is not possible to predict their collection efficiency, either on an overall mass basis or by particle size. Theoretical models to predict pressure drop are presently unreliable since they have not been calibrated with data. In contrast, efficiency and pressure drop models are available for most other particle collectors, such as cyclones, venturi scrubbers and electrostatic precipitators. The objective of this study is to provide pilot plant data for collection efficiency and pressure drop of pulse-jet fabric filters. The data will then be used to develop and calibrate models that will be able to predict pressure drop and collection efficiency for particles of various sizes. The models will be used in experiments to determine and verify optimum pulse-jet filter design and operating characteristics. Results form this study will be valuable to regulators concerned with estimating particle size and concentration from industrial sources that pass through pulse-jet filters. The results will also be useful to manufacturers and users of control equipment concerned with predetermining whether a proposed collector will meet emission regulations. To the extent that these studies will foster more effective, more reliable and lower-cost pollution control, filter users and the general public will also benefit.

Title:
New Approaches in Evaluating Environmental Impact in Marine Ecosystems

Principal Investigator:
Richard Levins

Institution:
Harvard School of Public Health

Project Period:
2 years

Budget:
(2 years) $296,596

EPA Project Number:
R 807713

Summary:

The objective of the research project is the development of a qualitative approach to environmental impact assessment in marine ecosystems. While not in an adversary position to a quantitative technique, the experimental approach will be directed toward an initial decipherability of ecosystem structure and function to allow for more selectivity in information quantification. Specific objectives are (1) the preparation of a casebook of network structures of several Atlantic Coast ecosystems from Virginia and Maryland, U.S.A. to the Davis Strait, Canada; (2) the determination of the effects of natural and manmade perturbations such as toxic chemicals, nutrient enrichment, overfishing and regulation on some of these networks depending on the availability of appropriate data sets; and (3) the development of the theory and applicability of two theoretical techniques (loop analysis and time averaging) for environmental impact assessment. Loop analysis is a network technique that can be used to predict parameter input to equilibrium communities in regard to directed changes, correlation and variability in the standing crops and turnover rates of a set of interconnected variables in a loop diagram. In time averaging, the assumption of equilibrium is relaxed. This technique combines concepts from the theory of stochastic processes with dynamical systems theory to analytically investigate systems which are described by functional equations. This allows for any sustained bounded motion in a system of many variables, thus making it more suitable for ecological systems which tend to be in constant and complex motion. Several ecosystem descriptions are complete or undergoing final revision. Theoretical predictions based on the analysis of two plankton communities have given good agreement with field data sets. This approach to environmental impact assessment is expected to be extremely cost effective.

Title:
Nitrification and Exchange of Dissolved N_2O and O_2 in Polluted Estuaries

Principal Investigators:
Michael B. McElroy
Steven C. Wofsy

Institution:
Harvard University

Project Period:
2 years

Budget:
(2 years) $238,904

EPA Project Number:
R 807081

Summary:

The program is based on field measurements in the Delaware and Potomac River estuaries. Longitudinal transects of these systems will be made for determination of dissolved nutrients (NH_4^+, NO_2^-, NO_3^-, PO_4^{-3}), organic N and P, and dissolved gases (N_2O, CH_4, CH_3Cl, O_2). The measurements of N_2O will be exploited to help quantify rates for nitrification of sewage NH_4^+ and the data will be systematically analyzed in order to elucidate the factors controlling nitrification in the rivers. Improved computer models for the nitrogen cycle on these rivers will be devised. Analysis of data for the Potomac has revealed nonlinear behavior of the nitrification rate in response to variations of freshwater flow into the tidal river. This phenomenon has proven to be consistent with a simple model for the growth kinetics of nitrifying bacteria. A paper describing these results has been submitted for publication. Preliminary analysis for the Delaware River has shown analogous behavior.

Title:
Effect of Preexisting Disease on Pulmonary Responses to Pollutant Particles and Gases

Principal Investigator:
Joseph D. Brain

Institution:
Harvard University

Project Period:
2 years

Budget:
(2 years) $265,516

EPA Project Number:
R 807082

Summary:

A major concern of environmental health policy is the "sensitive individual." Most animal research has focused on young, normal, healthy individuals, but there is growing concern for those that are unusually responsive, very young or very old, or those individuals with preexisting pulmonary disease. It is the purpose of the study to produce and characterize models of lung fibrosis and chronic bronchitis in rodents. The difference in the retention of toxic particles in these populations of animals and that of controlled populations will be measured. Both the amount and distribution of particles deposited will be quantified as will short-term and long-term clearance of particles using both radioactive and magnetic tracer techniques. Also examined will be the effects of acute and chronic exposure to nitrogen dioxide and ammonium sulfate on animals with preexisting lung disease. Using measurements of respiratory mechanics, determination as to whether the presence of disease causes measurable shifts in the dose repsonse curves for acute exposures to these agents will be ascertained. Of particular interest will be whether the no-measurable effect level for pollutant gases and particles is altered by lung disease. Using morphological and morphometric techniques, an examination will be conducted to establish whether chronic subacute exposures to nitrogen dioxide and ammonium sulfate influence the pathogenesis and progression of lung fibrosis and bronchitis.

Title:

Integrated Swine Waste Management System for Pollution Control and Energy and Food Production

Principal Investigator:
Ping-yi Yang

Institution:
University of Hawaii

Project Period:
2 years

Budget:
(2 years) $77,042

EPA Project Number:
R 808056

Summary:

The objectives of the project are (1) to develop necessary criteria for an effective, low-cost anaerobic digestion system with sludge recycle and raceway-algae-bacteria-growth system for swine waste and (2) to develop an improved, stable integrated swine waste management system for pollution control as well as energy and food production. Experimental data on a 20-m^3 anaerobic rubber digester (plug-flow) and a pilot algae-bacteria growing raceway (472x120x51 cm^3) will be used as a guideline to install, construct and operate an integrated swine waste management system at the Hawaii Institute of Tropical Agriculture and Human Resources. Optimal design and operation criteria will be determined and recommended (such as loading rate; sludge wastage rate and concentration of sludge recycle for anaerobic digester; nitrogen loading rate; mixing rate; hydraulic retention time; and maximum NH_4-N concentration for algae-bacteria-raceway system). At this time, a 20-m^3 rubber digester has been modified, installed, and operated at a pig farm on Oahu. Recycling part of the settled digester sludge has been practiced. This is expected to decrease reactor or digester volume and increase the manure loading rate. Also, a pilot raceway for algae-bacteria-growth system has been installed and operated at the University of Hawaii. This system is designed to investigate and evaluate the operational performance and kinetic behavior of algae-aerobic bacteria-growth and substrate (NH_4-N and COD) removal by feeding anaerobically digested effluent. Currently, an integrated swine waste management system is being constructed and is expected to be in operation March 1981.

Title:
Effects of Specific Radiofrequency Radiations on Human Cells

Principal Investigator:
S. K. Dutta

Institution:
Howard University

Project Period:
2 years

Budget:
(2 years) $116,975

EPA Project Number:
R 807490

Summary:

The primary objective of this project is to increase the information on the cellular effects of selective radiofrequency and microwave radiation by using standard human cell lines in culture. Evidence presented by other researchers has clearly shown cellular effects at particular frequency and power "windows," i.e., a limit range of values demonstrating positive effects. This project proposes to initially use a human neuroblastoma cell line AG 2202 or IMR-32 to scan for possible frequency and power "windows" using a fast and simple $^{45}Ca^{++}$ efflux technique that has been developed and used successfully in previous research experiments. Frequency and power density (windows) values will be established for neuroblastoma cells in culture. These values will be used to examine the response of other human cell lines (such as IMR-90) that are established from normal fibroblast tissue. Another phase of the project will examine the effects of short- and long-term exposure of normal skin primary explant in culture taken from human cadavers. These examinations are to investigate any possible effects on body tissue as opposed to individual cells. Experiments will be conducted with specific attention to the effects of microwave-induced heating so as to examine other possible causes of microwave-induced biological changes.

Title:

The Investigation and Demonstration of a Hydroperm Microfiltration System for the Treatment of Electroplating Wastewater Effluents

Principal Investigator:
H. L. Liu

Institution:
Hydronautics, Incorporated

Project Period:
1 year

Budget:
(1 year) $84,495

EPA Project Number:
R 807503

Summary:

The purpose of the program is to demonstrate onsite, the economic and technical viability of Hydroperm crossflow microfiltration for virtually complete removal of all suspended solids and toxic heavy metals from electroplating wastewater so as to produce water suitable for reuse and to minimize sludge handling problems. The electroplating industry produces a large volume (~2.5 billion gpd) of wastewaters which contain primarily four common toxic heavy metals (chromium, zinc, copper and nickel) as well as lead, iron, cadmium, tin, silver and others that amount to approximately 140,000,000 lbs/year. Current treatment technology generally involves conversion of the heavy metals into an insoluble hydroxide form, which is frequently hydrous, difficult to settle, requires large areas for conventional settling processes and involves hydrous sludge which is difficult to dewater and costly to transport. The application of Hydroperm microfiltration should have the following advantages: (1) virtually complete removal of suspended solids, including metal hydroxides in wastewater; (2) production of a sludge containing minimum water content with consequent reduction in handling cost; (3) production of a water product suitable for reuse in the rinsing (dragout) operation (an economic benefit); (4) minimization of space requirements; (5) reduction in capital and operating costs; (6) ready achievement of the TSS criterion established by environmental regulations; (7) reduction of industry cost resulting from imposition of environmental regulations; and (8) the provision of an effective modular treatment technology for small and large captive job operations.

Title:
Resistance of Algal Spores to Herbicide Treatments

Principal Investigator:
Jerome R. Cain

Institution:
Illinois State
University

Project Period:
2 years

Budget:
(2 years) $62,213

EPA Project Number:
R 807452

Summary:

The primary objective of this project is to determine if algal spores provide a means by which algae can resist elimination by chemical herbicides. The toxicity of many herbicides to the vegetative phases of many algal species has been studied. However, many algae, in addition to vegetative phases, produce spores as part of their sexual or asexual life cycles. No previous studies have examined the toxicity of herbicides to spore stages of algal life histories. This study is utilizing *Chlamydomonas moewusii*, a common freshwater alga, as a tool organism for the source of spores. Both the vegetative phase and spore phase of this alga's life cycle are being subjected to a range of concentrations of 22 different herbicides, which represent most of the chemical classes of herbicides in common usage. The ability of each herbicide to arrest growth of the vegetative phase is being evaluated. Also, the ability of each herbicide to prevent spore development and/or germination is bieng studied. Comparisons of the results obtained will determine if vegetative and spore stages differ in their sensitivity to various herbicides, and which ones are the most toxic to each phase.

Title:
Urine as a Biological Index of Exposure to Airborne Asbestos

Principal Investigator:
William Hallenbeck

Institution:
University of Illinois

Project Period:
1 year

Budget:
(1 year) $45,022

EPA Project Number:
R 807743

Summary:

The objectives of the project are: (1) to determine the best state-of-the-art methodology for preparing air and urine samples for analysis by transmission electron microscopy; (2) to determine if asbestos can be detected in the urines of workers chronically exposed to elevated levels of airborne asbestos and compare any positive findings to those derived from an appropriate control group; and (3) to characterize the airborne asbestos in the workers' environment. Characterization will be in terms of light microscopic analyses, TEM, fiber and mass concentrations, and length/diameter distributions. A group of 10 to 15 employees working in asbestos-exposed conditions will be studied at a Chicago factory which produces tars and sealers containing asbestos. Urine samples and personal air samples will be collected during the work shift. Detailed comparisons of the direct transfer and drop samples preparation techniques are being carried out in order to select the best methodology. Negotiations with factory officials concerning the schedule of air and urine sampling are in progress.

Title:

Limitations and Energetics of Hydrophobic (Nonpolar) Sorption of Organic Compounds by Soils and Sediments

Principal Investigator:
John J. Hassett

Institution:
University of Illinois

Project Period:
2 years

Budget:
(2 years) $144,438

EPA Project Number:
R 808417

Summary:

The objectives of the research project are to determine (1) the minimum K_{ow} value and/or the maximum water solubility (critical values) at which hydrophobic sorption becomes the dominant process independent of sorbent properties; (2) the sorbate and sorbent properties that control the type of sorption that dominates for compounds with water solubilities greater than or K_{ow} values less than the critical values; and (3) the free energies, enthalpies and entropies of sorption for a homologous series of polynuclear aromatic hydrocarbons, and other compounds with soils and sediments. Sorption isotherms will be conducted on a wide variety of compounds with water solubilities of approximately 100 ppm and above. Sorption of these compounds will be tested on selected soils and sediments that represent a wide range in sorbent properties, particularly a range in organic carbon to montmorillonite ratios. Results of these studies combined with existing data in the literature should facilitate definition of "critical values" for K_{ow} and S beyond which hydrophobic sorption would be expected to occur regardless of sorbent properties. The second objective will be approached by selecting specific compounds below the critical values established in objective one for strict hydrophobic sorption. Sorption of these compounds will be studied over a wide range of soils and sediments to further examine those sorbent properties that control the dominant type of sorption. Sorption constants will be determined from batch equilibrium sorption experiments carried out at constant temperature in temperature controlled shaking water baths. High performance liquid chromatography (HPLC) with variable wavelength UV-Vis and/or fluorescence detectors will be used.

Title:
The Development and Application of Factor Analysis to Urban Aerosol Source Resolution

Principal Investigator:
P. K. Hopke

Institution:
University of Illinois

Project Period:
2 years

Budget:
(2 years) $102,775

EPA Project Number:
R 808229

Summary:

This research explores the ability of target transformation factor analysis to perform quantitative aerosol source resolutions for both real and created data sets. Actual urban aerosol composition data from St. Louis, Missouri and Denver, Colorado will be analyzed. Additionally, data sets representative of airborne particle composition data will be created and analyzed to test the ability of the procedure to reproduce the original data including source composition and emission rates. These studies will clarify the strengths and limitations of factor analysis in source apportionment of urban aerosols. This project will yield a procedure that can be applied to development of cost-effective particle control strategies based on source resolution.

Title:
Mean Wind and Stability Over Complex Terrain of Moderate Relief on the High Plains: Analysis of Data Taken Around Colstrip, Montana

Principal Investigator:
Ronald Dobosy

Institution:
Iowa State University

Project Period:
1 year

Budget:
(1 year) $31,980

EPA Project Number:
R 808318

Summary:

Correct specification of an elective transporting wind and a general stability class is essential for operating air quality models. In complex terrain, finding these parameters can be difficult. Preliminary analysis of data taken around Colstrip, Montana, however, suggests a relatively simple characterization of mean winds and stability at least to first order if the terrain has moderate relief, as on the High Plains. It is proposed that in such terrain, for a substantial part of the year, daytime wind and stability structure resembles that over homogeneous terrain, while nighttime flow follows terrain drainage or channeling patterns up to at least 100m above the ground. This hypothesis will be tested through careful analysis of several years of Colstrip tower and surface data including a week of pibial winds and tethered balloon temperature profiles. Interpolated synoptic-scale flow will be compared with pibal and tower winds to test the first hypothesis statement. Prevailing stable flow up to 145m above Colstrip will be compared to terrain patterns to test the second statement. Factual information gained from this work will help in the design of efficient measurement programs for environmental impact assessments in moderate relief terrain.

Title:
Iowa Community Water Blood-Pressure Project

Principal Investigator:
Paul R. Pomrehn

Institution:
University of Iowa

Project Period:
2 years

Budget:
(2 years) $201,369

EPA Project Number:
R 807217

Summary:

A survey measuring blood pressure, heart rate, height, weight and triceps shinfold in 1,500 ten-, eleven- and twelve-year old children in eight (8) Iowa communities will be conducted to determine if: (1) water sodium levels relate to mean blood pressure; (2) the water sodium level-mean blood pressure relationship interacts with water hardness; and (3) family history of hypertension enhances the water sodium level-mean blood pressure relationship. Communities were selected for diversity in water sodium levels (from over 200 Mg/l to less than 10 Mg/l variation) and demographic similarity. All measurements will be made by trained observers using standardized techniques. Samples of water from participants' homes will be analyzed for Na^+, K^+, Ca^{++}, Mg^{++} and $CaCO_3$. Further, a subset of parents will be selected and home visits conducted to measure blood pressure, heart rate, height, weight, and triceps shinfold. Parents will be stratified into three groups: high, middle, and low according to blood pressure. Blood pressure of children whose parents are in high, medium, and low groups will be compared contrasting high water sodium level communities with low water sodium level communities. This contrast will test if the pressor response to water sodium levels is enhanced by family history of hypertension. Results will be intrrepreted with consideration given to health effects of various water sodium levels and water treatment practices.

Title:
Implications of the Mode of Action of Chlorine Dioxide on Virus

Principal Investigator:
Vincent P. Olivieri

Institution:
Johns Hopkins University

Project Period:
2 years

Budget:
(2 years) $385,900

EPA Project Number:
R 807747

Summary:

The overall objective of the proposed project is to evaluate the implications of the mechanism of inactivation of viruses with chlorine dioxide. The viruses employed will be the bacterial viruses f2 and ØX174, poliovirus, and swine enterovirus. The specific objectives are: (1) to determine the biological function of viral nucleic acid and protein after treatment with chlorine dioxide; (2) to evaluate the structural integrity of the virion after treatment; (3) to determine the fate and stability of the treated viral components in the aquatic environment; and (4) to evaluate the infectivity of treated virus in systems other than cell monolayers. The approach employed will be to follow the biological activity of the virus during and after treatment with chlorine dioxide. The initial phases of the study will be to determine the ability of viral protein and nucleic acid to function and to evaluate structural integrity of the treated virus. If nucleic acids remain infectious after exposure to levels of chlorine dioxide commonly employed in water and wastewater treatment, the survival of these preparations will be evaluated in aquatic systems. Also, the infectivity of the chlorine dioxide treated virus will be evaluated in tissue culture and animals to determine whether chlorine dioxide inactivated virus can produce infection in animals.

Title:

Coagulation/Precipitation of Natural Organic Substances in Water Treatment

Principal Investigator:
Charles R. O'Melia

Institution:
Johns Hopkins University

Project Period:
2 years

Budget:
(2 years) $142,749

EPA Project Number:
R 808104

Summary:

The goals of this project are (1) to characterize the abilities of coagulant chemicals to remove organic matter from water and determine certain properties of the organics remaining in solution after treatment; (2) to note the chemical conditions affecting the ability of metal salts to remove natural organic matter, including the effects of competing pollutants in the water, the use of preformed solid phases, and the strength and composition of coagulant stock solutions; (3) to determine the effects of reactor design on the reactions induced by metal salts and organic polymers, involving such parameters as mixing intensity and hydraulic reaction time in rapid mix and flocculation units; and (4) to combine this information with other veritable data (i.e.,chemical coagulants, natural organic substances, and treatment reactors) into a treatment system for water supplies containing both soluble organic substances and particulate matter (turbidity).

Title:
Wet Oxidation of the Wastewater from Coal Gasification Installations

Principal Investigator:
O. J. Hahn

Institution:
University of Kentucky

Project Period:
2 years

Budget:
(2 years) $245,275

EPA Project Number:
R 807666

Summary:

The objectives of the proposed research are to demonstrate with the University of Kentucky's half ton per day moving bed coal gasifier and small laboratory apparata: (1) that wet oxidation of organic and inorganic material in the wastewater from gasifiers is technically feasible and economically attractive; (2) to determine the kinetic parameters controlling the process; and (3) to find possible catalysts to accelerate the reaction. The feasibility of wet oxidation to treat wastewaters has been demonstrated in a small pressure vessel. However, no reaction data has been determined. In this project the wastewater is generated with an existing and proven half ton per day coal gasifier. The initial reaction parameters and catalysts will be determined in small autoclave systems. In the second year a small test loop will be incorporated into the existing gasifier to confirm reaction parameters and investigate buildup of trace elements in the treated and recirculated wastewater.

Title:
Carcinogen Mechanism—Interferon Induction Inhibition

Principal Investigator:
Gerald Sonnenfeld

Institution:
University of Louisville

Project Period:
2 years

Budget:
(2 years) $120,921

EPA Project Number:
R 807619

Summary:

The long-term goals of this project are to study and delineate the effect of carcinogens on interferon induction and action and to develop the inhibition of the interferon induction system as a reliable assay for the carcinogenic potential of chemicals. The number of chemicals tested in the system will be expanded to include several known carcinogens, their rarely- or non-carcinogenic analogs, and non-carcinogens. Determination if the inhibition of interferon induction can discriminate among chemicals on the basis of carcinogenic potential will hopefully be established. An attempt will be made to correlate the results of the inhibition of the interferon induction system with other assays for chemical carcinogenicity (e.g., Ames test, Comptest) in efforts to establish the inhibition of the interferon induction system as a reliable assay. Continued discrimination among chemicals may lead to development of a human *in vitro* screening assay for carcinogens based on the inhibition of interferon induction.

Title:
Utilization of Peat in Onsite Wastewater Disposal Systems

Principal Investigator:
Chet A. Rock

Institution:
University of Maine

Project Period:
2 years

Budget:
(2 years) ***$45,789***

EPA Project Number:
R 808069

Summary:

The objective of this project is to develop an inexpensive alternative on-site waste disposal system that can be utilized in areas that do not have acceptable soils for conventional drainfield systems. The project will define and establish the design parameters needed to construct and operate a practical peat adsorption bed receiving septic tank effluent. The springboard for the project is the very encouraging results obtained by Brooks (1980), who monitored a peat adsorption field. The project is proposed in two phases: (1) laboratory studies and (2) field site monitoring. Initial efforts will be centered on laboratory evaluation of design parameters and peat evaluation, with resumption of additional monitoring of Brooks' original installation. A second peat adsorption system will be designed and installed during the 1981 construction season based on the results obtained by Brooks. After sufficient information is gathered in the laboratory through studies of peat removal efficiencies under a variety of conditions, a third system may be constructed in 1982. Peat will also be evaluated as an alternative to sand in the raised bed (mound) system and an experimental system will be installed and monitored.

Title:
Heterogeneous Catalytic Oxidation of Model Chlorinated Hydrocarbons

Principal Investigator:
Michael P. Manning

Institution:
Massachusetts
Institute of
Technology

Project Period:
2 years

Budget:
(2 years) $167,970

EPA Project Number:
R 808296

Summary:

[illegible] objectives of this project are: (1) to measure the kinetics of heterogeneous oxidation of three sets of model compounds on three catalysts, supported V_2O_5, Cr_2O_3, and an oxyhydrochlorination catalyst; (2) to correlate the measured rates and determine rate expressions for the oxidation of the chlorinated paraffins, olefins, and aromatic compounds as a function of temperature and reactant concentrations; and (3) to assess the effect of nitrogen and sulfur as a potential catalyst poison by measuring the rates of catalytic oxidation in the presence of SO_2 and NO_2. The kinetics of heterogeneous catalytic oxidation of model paraffinic, olefinic and aromatic chlorinated compounds will be measured in a tubular flow reactor. Inlet and outlet compositions will be measured by gas chromatography. Support vanadia and chromia catalysts and an oxyhydrochlorination catalyst will be examined. The effects of nitrogen and sulfur as catalyst poisons will be tested by introduction of NO_2 and SO_2 into the reactor feed.

Title:
Thermal Destruction Kinetics of Chlorinated Hydrocarbons

Principal Investigator:
Selim Senkan

Institution:
Massachusetts Institute of Technology

Project Period:
2 years

Budget:
(2 years) $198,117

EPA Project Number:
R 808314

Summary:

Controlled thermal destruction of chlorinated hydrocarbons (CHC) is an attractive method in the ultimate disposal of these hazardous chemical wastes. The combustion principle involved in CHC oxidation is currently at a very primitive state, and is inadequate for the design and operation of reliable incinerators. The combustion characteristics of various CHC compounds are being studied in order to develop veritable data on the engineering and scientific principles involved in hazardous chemical waste incinerator design. Premixed flame studies will be directed towards developing an understanding of the kinetics of flame destruction of three classes of chlorinated hydrocarbons; chloromethanes, chloroethylenes, and chlorobenzenes. Experiments will be conducted using two burner systems: a flat-flame burner and a Bunsen burner, with the bulk of the studies being conducted with the former. CHC destruction rates will be studied as a function of changing Cl content and the nature of the CHC compound, temperature, excess air, and auxiliary fuel (CH_4) use. Single CHC compounds as well as their mixtures will be investigated. In parallel with this experimental work, theoretical studies will also be undertaken to model the flame structure.

Title:

Investigation of the Potential for Nitrosamine Production on Activated Carbon in Treatment of Drinking Water

Principal Investigator:
Francis A. DiGiano

Institution:
University of Massachusetts

Project Period:
2 years

Budget:
(2 years) $133,425

EPA Project Number:
R 807537

Summary:

The regulation concerning use of granular activated carbon (GAC) in water work installations has been carefully scrutinized by the industry. There is concern regarding possible contaminant production on the GAC surface. The proposed research postulates that N-nitrosamines, which have been shown to be carcinogenic, may in fact be produced on GAC surfaces through reactions of nitrite and amines or other organic precursors. Nitrite is derived from microbial conversion of ammonia to nitrate. The probability of nitrosamine production on GAC surfaces, although remote, has not been refuted by scientific research. Nonetheless, nitrosamine production has been noted on de-ionizing resins. Through laboratory study, this project will investigate and characterize the microbial and catalytic contamination caused by nitrosamine production on GAC surfaces.

Title:

Cooperative Agreement to Study the Effects of Municipal Sludge (Milorganite) on the Structure and Function of Two Grassland Ecosystems

Principal Investigator:
Gary W. Barrett

Institution:
Miami University

Project Period:
2 years

Budget:
(2 years) $46,529

EPA Project Number:
R 807370

Summary:

The major objectives of this investigation are (a) to measure and evaluate the effects of chronic sludge application on several different levels of biological organization; (b) to compare two different grassland community types treated in an identical manner; (c) to analyze different structural (e.g., biomass and species diversity) and functional (e.g., energy flow and community resilience) ecosystem parameters which may be indicative of total systems response; (d) to evaluate sludge effects via ecosystem food chains on a natural small mammal population (*Microtus pennsylvanicus*) functioning within these community types; and (e) to develop a feasible ecosystem approach to testing stressors on intact ecosystems, including the identification of "white mice" field parameters. Additional questions to be answered include: (1) Are older, more diverse ecosystems better able to assimilate and recycle nutrient input than younger, less diverse ecosystems; (2) Will nutrient input produce trophic level differences between the two community types; (3) Will small mammal reproduction and/or growth rates be significantly affected either between control and treatment plots or between old-field community types; (4) Which type of ecosystem will manifest the greater net energy benefit to mankind when treated experimentally with sewage treatment sludge; and (5) Will sludge research studies serve as a model to provide knowledge for interface management between man-made and natural ecosystems?

Title:
Assessment of Energy—Related Toxicant Stress in an Engineered Ecosystem

Principal Investigator:
Robert H. Boling, Jr.

Institution:
Michigan State University

Project Period:
1 year

Budget:
(1 year) $351,095

EPA Project Number:
R 807642

Summary:

The basic objectives of this project are to develop expertise and refine an experimental facility to evaluate the relevancy of toxicant testing protocols (e.g., microcosms and other laboratory toxicity tests) to field conditions. The facility (two 3000-foot long outdoor recirculatory channels) will be stressed by moderate levels (8 ppm) of p-cresol in controlled 24- and 48-hour tests and the results compared to laboratory predictions.

Title:
Neurotoxicity Screening Assay

Principal Investigator:
Roger E. Davis

Institution:
University of
Michigan

Project Period:
2 years

Budget:
(2 years) $72,518

EPA Project Number:
R 808064

Summary:

The objective of this study is to further develop a laboratory protocol for assessing the acute neurotoxicity of chemical pollutants *in vivo*. The thesis is that developing or regenerating neurons are more sensitive indicators of toxic effects than mature neurons. A toxicant might inhibit neuronal growth at concentrations which have no apparent effect on the maintenance of function in mature neurons. Regenerating optic nerve fibers in the goldfish provide an advantageous, experimental preparation for assessing toxic effects of substances on neuronal development. Regeneration is induced by crushing the nerve. The subsequent recovery of vision is measured, as an index of the rate of regeneration, by a non-invasive behavioral technique which permits sequential testing in individuals. Experiments with alkaloid neurotoxins confirm that the protocol can be used to obtain concentration percent response data for estimating the relative potency of neurotoxicants *in vivo*. The first three months of the project has seen the completion of an improved aquarium facility and development of new electronic apparatus for conditioning and testing a respiratory ventilation response to light. The facility permits housing up to eighty fish in individual containers, with an automatically regulated supply of fresh, dechlorinated, pH and temperature-controlled water. Methods of recording respiratory response, visual stimulus presentation and running the test trials have been extensively revised.

Title:
Synergistic Effects of Heavy Metals and Macronutrients on Toxicant Bioaccumulation

Principal Investigator:
Linda Sicko Goad

Institution:
University of Michigan

Project Period:
2 years

Budget:
(2 years) $73,613

EPA Project Number:
R 807516

Summary:

The primary objectives of this project are: (1) to determine the importance of polyphosphate formation in allowing the dispersal and dominance of nuisance producing phytoplankton populations in aquatic systems; (2) to determine if polyphosphate formation is an important mechanism in the uptake and biological transport and bioaccumulation of toxic heavy metals; and (3) to investigate the synergistic effects of combined phosphorus and heavy metal loadings. The approach to be used in determining the importance of polyphosphate as a toxicant dispersal mechanism will consist of both field and laboratory studies. The first year of the project will be devoted to assessing the importance of polyphosphate in natural phytoplankton assemblages in Saginaw Bay and southern Lake Huron. Previous demonstrations have shown (1) that certain phytoplankton populations were exiting the bay and surviving transport to southern Lake Huron; (2) that many of these populations had polyphosphate accumulations in them; and (3) that several of the populations had lead (Pb) sequestered in the polyphosphate bodies. Evaluations will be made by microscopical methodology to indicate the frequency and spatial/temporal occurrence of polyphosphate in natural assemblages in both Saginaw Bay and southern Lake Huron. These data will enable assessment of two important factors: (1) Is there a significant biological/intracellular transport of phosphate from highly eutrophic areas to less directly affected parts of the Great Lakes ecosystem; and (2) Did the reduction (approximately 50%) in phosphorus loading to Saginaw Bay result in decreased stored phosphates? The second phase of the research will address the following questions by a series of laboratory experiments: (1) Does polyphosphate serve as a phosphorus storage form that allows survival of cells under conditions of low available phosphorus; (2) Can polyphosphate be released upon death of transported species and subsequently stimulate growth of other species; and (3) If lead (and possible other heavy metals) are sequestered in polyphosphate, can it be mobilized within the cell when polyphosphate is degraded enzymatically?

Title:
Conservative Ion Effects on Phytoplankton

Principal Investigator:
Eugene F. Stoermer

Institution:
University of Michigan

Project Period:
2 years

Budget:
(2 years) $117,499

EPA Project Number:
R 807547

Summary:

Long-term records of phytoplankton succession in the Great Lakes suggest that undesirable modifications of the flora result from low-level salinification. Preliminary evidence suggests that increased concentrations of certain conservative ions give certain phytoplankton populations competitive advantage in uptake of critical or limiting nutrients. This problem is particularly crucial in oligotrophic, long residence systems, such as the Great Lakes. It is proposed that three types of experiments be conducted: (1) determination of the effects of low-level perturbations of conservative ion concentration on the growth rate of specific phytoplanktons; (2) studies using naturally occurring assemblages to determine if low-level perturbations of conservative ion concentration causes modifications of assemblage composition; and (3) retrospective studies of a small lake system which has a known history of salinification to determine if effects generated in the laboratory occur in a complete ecosystem. The results of this study should quantify the potential for environmental damage from low-level salinification and suggest appropriate levels of control.

Title:

Enhancement of the Removal of Humic Substances and THM Precursors from Water Supplies

Principal Investigator:
Walter J. Weber, Jr.

Institution:
University of Michigan

Project Period:
2 years

Budget:
(2 years) $163,400

EPA Project Number:
R 807359

Summary:

The ubiquity of humic acids in drinking water supplies, coupled with their significance as trihalomethane precursors, dictates the need for development of efficient and cost-effective general methods for their removal in water treatment operations. The findings of exploratory research indicate that the adsorptive affinity of activated carbon for humic acids is markedly dependent upon the presence and concentrations of several common constituents of water supplies, specifically Ca^{2+}, Mg^{2+}, and OCl^-. These findings also suggest that carbon treatment of water for humic acids removal might be rendered more efficient and cost-effective in many instances by addition of relatively common and inexpensive reagents. The specific objective of the proposed research is to increase the amount of quantifiable data relative to adsorption phenomena, and to explore the possible enhancement of humic acids removal by optimization of the carbon adsorption process. The exploratory work gives further evidence that both bioconversion and catalytic conversion of certain fractions of humic acids may be operative in practical applications of activated carbon in water treatment. Characterization and quantification of the biophysico-chemical dynamics of humic substance interactions with activated carbon constitutes a secondary objective of the proposed research.

Title:
The Development of Particulate Impact Mass Spectrometry for Use in the Analysis of Nonvolatile Trace Pollutants in Water and Other Systems

Principal Investigator:
Frank T. Greene

Institution:
Midwest Research Institute

Project Period:
2 years

Budget:
(2 years) $124,193

EPA Project Number:
R 807636

Summary:

The objective of the proposed project is the development of a technique for the analysis of traces of nonvolatile organic materials in water. The major emphasis of the project will be on the characterization of particulate impact mass spectrometry (PIMS). Experiments will be carried out to characterize the PIMS volatilization process and to determine the degree of universatility of the technique. Some research will also be directed toward increasing the sensitivity of the technique.

Title:

Amino Acid Incorporation and RNA/DNA Ratio as Rapid Methods for Toxicity Tests

Principal Investigator:
Ira R. Adelman

Institution:
University of Minnesota

Project Period:
2 years

Budget:
(1 year) $107,357

EPA Project Number:
R 807360

Summary:

The project has been undertaken to determine if amino acid incorporation and/or RNA/DNA ratios will be affected by toxicants within 96 hours and will predict effects on growth resulting from chronic exposures. Larval stage fathead minnows will be exposed to different toxicants in a series of tests. During the first 4 days of exposure, fish will be sampled from each toxicant concentration, RNA and DNA content of whole fish will be analyzed and ^{14}C-glycine incorporation by axial musculature will be determined *in vitro*. At 30 days exposure, the remaining fish will be measured and weighed. Toxicant concentrations affecting actual growth at 30 days will be compared to those affecting the short-term indicators. At present, methodology for the glycine incorporation experiment is being perfected. Uptake is linear up to at least 4 hours.

Title:
The Fate of PNAs During Aqueous ClO_2 Treatment

Principal Investigator:
Robert M. Carlson

Institution:
University of Minnesota

Project Period:
2 years

Budget:
(2 years) $139,113

EPA Project Number:
R 807455

Summary:

The rates and product distributions will be determined for the reactions of polycyclic aromatic compounds (PNA) in aqueous chlorine dioxide (ClO_2) solutions under conditions that will be encountered in industrial and municipal water treatment facilities. The rate data obtained for each aromatic compound will be described by equations which would allow the prediction of the rates of reaction of the compound under any specific set of conditions. The products identified will be synthesized and made available for biological testing. Moreover, these pure compounds will represent standards for quantitation as well as for evaluation of environmental samples by gas chromatography/mass spectrometry (GC/MS). The results obtained in this project will be directly compared to those obtained in a related project in which chlorine is used as the oxidant. This comparison will aid in the assessment of the environmental implications of using chlorine dioxide in place of chlorine or in conjunction with chlorine in water treatment facilities. The proposed studies will include the determination of effects of the chlorite (ClO_2^-), chlorate (ClO_3^-) and/or chloride (Cl^-) ions as well as the effects of chlorine (as HOCl, Cl_2, and/or OCl^-) upon the rates and product distributions of aqueous chlorine dioxide reactions. The information derived from these experiments will provide insight into the presently unknown source of chlorine-containing compounds in some reported chlorine dioxide reactions. It will also be of use in optimizing chlorine dioxide generation and disinfection conditions to minimize the production of biologically harmful compounds.

Title:

Clinical Approach to Quantitating Formaldehyde Effects in Human Blood

Principal Investigators:
V. F. Garry
Alfred D. Barksdale

Institution:
University of Minnesota

Project Period:
1 year

Budget:
(1 year) $29,758

EPA Project Number:
R 807220

Summary:

The first purpose of this study is to identify laboratory methodology and to determine the feasibility of its use for direct quantitation of formaldehyde effects in man. To meet this primary objective, chemical and bioassay methods as applied to human peripheral blood will be considered. The sensitivity, specificity and normal range of the assays, as well as their clinical applicability in routine practice will be evaluated. The second purpose is that of identifying the chemical, biochemical and immunological mechanisms by which formaldehyde directly or indirectly produces a diseased state. Because of the complexity of the chemical, biochemical and cellular pathways which are potential routes to the diseased state associated with formaldehyde exposure, parallel, multidisciplinary studies of formaldehyde effects on peripheral blood constituents are expected to provide the most thorough and rapid analysis of the problem. The experimental approach entails (1) the documentation of effects of formaldehyde on the formed elements of blood in order to assess the role of formaldehyde in producing a diseased state; (2) the development of an assay for formaldehyde in human peripheral blood, as a first step in evaluating the effects of formaldehyde on the immune system, leucocyte chemotaxis, or possible cellular damage; and (3) a systematic investigation of classical clinical and biochemical methods as potential means for formaldehyde detection. Benefits of the program include the development of a standard assay for formaldehyde in blood, and the production of information concerning the underlying molecular and cellular events attendant to formaldehyde poisoning.

Title:

A Fundamental Study of the Mechanics of Aerosol Sampling and Sampling Inlets

Principal Investigator:
Benjamin Y. H. Liu

Institution:
University of Minnesota

Project Period:
2 years

Budget:
(1 year) $99,787

EPA Project Number:
R 807471

Summary:

The primary objective of this research program is to make a fundamental study of the mechanics of aerosol sampling and the performance of sampling inlets for atmospheric aerosol sampling. The characteristics of various sampling inlets will be determined as a function of wind speed and sampling flowrate. Both theoretical and experimental studies will be undertaken; the ultimate objective is to gain fundamental information on the particle and fluid mechanics involved so that inlets of high efficiency can be designed. The output of the program will be a set of design guidelines which can be used to design inlets of high efficiency for atmospheric aerosol sampling. In addition, a series of sampling inlets of different flowrates will be designed, tested and carefully characterized in the wind tunnel so that they can be used as general purpose inlets for atmospheric aerosol sampling. The theoretical studies will be made by first determining the flow fields near the entrance to the inlet, and then determining the particle trajectories through these flow fields by solving the particle equations of motion. The flow fields used will include the potential flow field similar to that used by Zebel and viscous flow fields obtained by solving the Navier-Stokes equations. The experimental studies will involve measuring the sampling efficiency of various aerosol inlets in a wind tunnel by means of monodisperse test aerosols generated by a vibrating orifice monodisperse aerosol generator. The computer program for calculating the flow fields and particle trajectories is in the process of development. Wind tunnel modifications are in progress. One inlet has been constructed and partially tested and another one is under design. A study on methods for sizing liquid aerosols to be used in the program has been completed and a finished paper describing this study has been submitted to a journal.

Title:

Application of Fourier Transform Mass Spectrometry to the Analysis of Organic Emission Samples

Principal Investigator:
Charles L. Wilkins

Institution:
University of Nebraska

Project Period:
2 years

Budget:
(2 years) $107,959

EPA Project Number:
R 807251

Summary:

The objective of this project is the development of a new GC-mass spectrometry tool of unparalleled speed, resolution, and sensitivity by means of continued development of the Fourier transform mass spectrometer. This spectrometer, which is used as an analytical tool, will provide capabilities for high speed, high sensitivity analysis of complex mixtures. This will be accomplished by design and installation of an interface between a capillary column gas chromatograph and a Fourier transform mass spectrometer. This combination is expected to allow realization of the enormous analytical potential of the recently-established Fourier transform mass spectrometric technique. One focus of the research will be the development of suitable hardware/software interfaces which permit computer-controlled electron impact and low pressure chemical ionization analysis to be carried out on the same samples in rapid sequence. The new techniques thus developed will be evaluated for practical analytical usefulness by analysis of representative mixtures containing polycyclic organic matter (POM) typical of organic stack emission samples. Quantitative comparisons of the results of such analysis will be made with results from benchmark measurements performed using the ultrahigh resolution MS-5076 mass spectrometer available in the laboratory.

Title:

Toxicity of Heavy Metals to Microbes and Microbe-Mediated Ecologic Processes: Effect of Chemical Form and Environmental Factors

Principal Investigator:
Guenther Stotzky

Institution:
New York University

Project Period:
2 years

Budget:
(2 years) $256,000

EPA Project Number:
R 808329

Summary:

The results of this study will provide more comprehensive knowledge of the interactions of heavy metals with the abiotic components of aquatic and terrestrial environments and of the effects of these interactions on the potential toxicity of heavy metals to microorganisms and microbe-mediated ecological processes. This data will provide a basis for establishing new criteria for environmental toxicants and will serve as a guide to research on ameliorating the toxic effects of these pollutants. The information derived from this research will be used to formulate ecological dosage 50% (ED_{50}) values, which can be used to set criteria. The ED_{50} concept has many applications. Its concept and formulation has three main advantages over setting of criteria based solely on LD_{50} values: (1) an ED_{50} reduction in one ecosystem can easily be correlated with an equivalent reduction in the *same* ecological process in a different ecosystem; (2) ED_{50} value reflects the response of spectrum of organisms to a common pollutant; and (3) an ED_{50} value enables a direct comparison of the relative fragility of different environments to the same pollutant. The implementation of ED_{50} values would provide a simple and effective means of regulator toxicants in terrestrial and aquatic ecosystems.

Title:
Effects of Exposure to Summertime Haze Episodes on the Health of Children

Principal Investigator:
Morton Lippmann

Institution:
New York University
Medical Center

Project Period:
1 year

Budget:
(1 year) $80,174

EPA Project Number:
R 807723

Summary:

The objectives of the proposed study are (1) to characterize ambient air pollutants, especially ozone and sulfuric acid, in a small, western Pennsylvania city; and (2) to characterize responses to these air pollutants among exposed children. There are no major local pollution sources nearby, but New York City is in the region most affected by large regional scale summertime pollution episodes which originate in the Ohio River Valley. The project will involve comparison of the pollutant levels measured during a Prolonged Elevated Pollution Episode (PEPE) and the population responses recorded in the health effects studies. The main feature of these analyses will be correspondences drawn between decreases in pulmonary function, such as in FEV$_{1.0}$, FEV$_{0.75}$, and FVC, and increases in air pollution. Confounding factors of disease and other physiological and environmental stresses will be considered from the results of the questionnaire analyses. At a minimum, population exposure estimates to the measured pollutants will be determined.

Title:

Electrosorption of Phenol on Activated Carbon from Dilute Aqueous Solutions

Principal Investigator:
Peter S. Fedkiw

Institution:
North Carolina
State University

Project Period:
2 years

Budget:
(2 years) $49,077

EPA Project Number:
R 808216

Summary:

The purpose of this project is to determine the technological feasibility of increasing the adsorption capacity of an activated carbon adsorber for dilute nonionic organic species by applying an electrical potential to the adsorbent. The applied potential, depending upon its magnitude, will either increase or decrease the free energy of adsorption. Adsorption under an imposed voltage is called electrosorption to distinguish it from adsorption in the absence of an applied electric field. The requisite characteristics of a successful electrosorption process are described. Control of the applied potential is crucial; too high (positive) or too low (negative) of an applied potential will cause unwanted faradaic oxidations and reductions, respectively. If the electrosorption process is reversible, a simple change of the polarity of the applied voltage to the carbon can be used to regenerate the bed. This electroregeneration should be compared to the more complex solvent or stream stripping procedures used for regeneration of conventional adsorbers. Removal of phenol from a dilute (less than 100 ppm) aqueous solution will be used as the model system. Phenol is one of the hazardous pollutants which is found in the condensate water from a coal gasification plant. The two principal objectives are: (1) to determine the equilibrium electrosorption isotherm of phenol on activated carbon; and (2) to evaluate the elution characteristics of a fixed-bed activated carbon electrosorber. The effects of temperature, pH, O_2 content, phenol concentration and applied voltage on the equilibrium electrosorption isotherm will be studied.

Title:
Modeling Observed Hydrocarbon Mixture Reactivity Effects

Principal Investigator:
Harvey E. Jeffries

Institution:
University of
North Carolina

Project Period:
2 years

Budget:
(2 years) $184,603

EPA Project Number:
R 807762

Summary:

The objective of this project is to make photochemical kinetics models (PKMs) for use in urban air shed simulation models (UASMs) that correctly represent the effects that occur when the composition of urban hydrocarbon (HC) mixtures change. Necessary sub-objectives are (1) to generate a systematic data base that describes experimentally the effects of changing HC composition in a readily modelable chamber using urban-like chemical and physical conditions; (2) to model these experiments with PKMs now in use; and (3) to reconcile the difference between the models and experiments by changing the models. The overall approach is a direct comparison of model and experiment. The methods to be used include: (1) computer simulations using existing PKMs; (2) mathematical and mechanistic analysis of cause-and-effect relationships in models; (3) dual smog chamber experiments in which identical conditions are used on both sides except for HC composition to directly and systematically demonstrate the effects of composition changes; and (4) subsequent remodeling with modified cause-and-effect relationships to improve predictions. The primary product will be better models for use in UASMs. Other products will include a magnetic tape archive of experiment results, EPA reports and journal publications in which experiments and model results will be discussed, and suggestions for expanding fundamental research in support of these models.

Title:

Effect of Various Nitrogen Sources and Management Practices on Water Quality of Cattle Pasture Surface and Subsurface Runoff Water

Principal Investigator:
R. W. Van Keuren

Institution:
Ohio Agricultural
Research and
Development Center

Project Period:
2 years

Budget:
(2 years) $127,179

EPA Project Number:
R 808098

Summary:

The purpose of this project is to determine the effect of several nitrogen sources and establish the best management practices for forage production and cattle grazing on hill lands in order to improve the quality of surface and subsurface runoff water. Beef cow and calf herds will be kept under three forage/livestock management systems. Each area will have instrumented watersheds. A wooded, central watershed will be monitored as a control (background level). Surface water runoff and sediment and subsurface water will be collected and measured. Subsamples will then be taken to determine the levels of mineral and organic nitrogen, phosphorus, and other chemical measurements of the water and sediment. Standard laboratory procedures will be used for making the above determinations. During 1981 the effect of summer grazing and winter feeding of cattle on water quality and sediment movement on two forage programs will be studied at two test sites. One test plot will have a moderate level of soil fertility and several fertilizer nitrogen sources for summer grass pasture, with winter feeding of large round bales in one of the four summer pastures. The other plot will have low soil fertility, continuously grazed summer pasture, no applied fertilizer, with large round bales fed into pasture during winter. Also, orchard grass-alfalfa and tall fescue-alfalfa fields were established in 1980 for a high soil fertility/grass legume study to be initiated in 1981.

Title:
Fate of Environmentally Induced DNA Damage in Human Fetal Tissues

Principal Investigator:
Steven M. D'Ambrosio

Institution:
Ohio State University

Project Period:
2 years

Budget:
(2 years) $173,237

EPA Project Number:
R 807693

Summary:

Previous studies have shown that: (a) alkylating agents and polyaromatic hydrocarbons (PAH) are potent transplacental carcinogens at least in the rodent animal models; (b) human epidemiological data indicate that over 90% of the cancers detected within the first ten years of life are thought to originate by exposure of the fetus to carcinogen; (c) DNA damage and repair (or lack of it) plays an important role in the initiation of cancer; (d) DNA repair changes during rodent embryogenesis; and (e) there appear to be differences in the level of repair, at least for ultraviolet radiation, in human cells derived from different organs of the same fetus. This research project proposes as its main objectives to: (a) determine the effects of environmental agents upon cells in culture derived from human fetal and embryonic tissues; and (b) understand the molecular events leading to the role of DNA damage and repair in the induction of human transplacental carcinogenesis in humans.

Title:
Disinfection of Giardia lamblia *Cysts with Chlorine, Chlorine Dioxide and Ozone*

Principal Investigator:
Otis J. Sproul

Institution:
Ohio State University
Research Foundation

Project Period:
2 years

Budget:
(2 years) $116,573

EPA Project Number:
R 808150

Summary:

The project will determine the inactivation kinetics of cysts of *Giardia lamblia* and *Naegleria gruberi*: using chlorine, chlorine dioxide and ozone as disinfectants. The effects of pH, contact time, cyst and disinfectant concentration, and temperature will be determined.

Title:

Organic Micropollutants in Urban Precipitation

Principal Investigator:
James F. Pankow

Institution:
Oregon Graduate Center

Project Period:
2 years

Budget:
(2 years) $115,317

EPA Project Number:
R 807239

Summary:

The objectives of this project are (1) to determine identities and concentrations of organic compounds present in precipitation falling in Portland, Oregon and Los Angeles, California; (2) to correlate these data with concentrations of inorganic constituents; (3) to determine the atmospheric levels of these organics during storm events; (4) to compare precipitation and atmospheric levels of the organics and determine the extent these same organics are scavenged from the atmosphere; and (5) to determine and describe the relative magnitudes of gas and aerosol scavenging. Both the dissolved and particulate organics will be concentrated from precipitation samples using resin adsorption and solvent extraction techniques. Gaseous and aerosol organics will be concentrated from atmospheric samples using resin traps and filters. Analysis of the organics will be carried out by GC/MS and that of the inorganics by inductively coupled organ plasma spectroscopy, ion chromatography, and potentiometry.

Title:

Evaluation of Chemical Receptor Models for Air Particulate Source Impact Assessment Using the PACs Data Set

Principal Investigator:
John A. Cooper

Institution:
Oregon Graduate
Center for Study and
Research

Project Period:
2 years

Budget:
(2 years) $140,500

EPA Project Number:
R 807375

Summary:

The primary objective of this program is to fundamentally study the mechanics of aerosol sampling and the performance of sampling inlets. The ultimate objective is to gain an understanding of the particle and fluid mechanics involved so that improved inlets of high efficiency can be developed and design guidelines established. A series of sampling inlets of different flowrates will also be developed and tested so that they can be used directly by researchers and monitoring agencies for atmospheric aerosol sampling and measurement.

Title:
Measurement of the Dry Deposition Velocity of Atmospheric Aerosol Particles

Principal Investigator:
Paul C. Katen

Institution:
Oregon State University

Project Period:
2 years

Budget:
(2 years) $219,252

EPA Project Number:
R 807902

Summary:

The basic objective of this proposal is to investigate, improve and compare measurements and the measuring instruments used for determining the dry deposition velocity (v_d) of atmospheric aerosol particles by the profile and eddy-correlation methods. Specifically, a PMS ASAS-300 A and a Royco 225 will be developed into sensors that can be used to make aerosol flux measurements by the eddy-correlation technique. These sensors will allow detailed v_d measurements (as small as 0.01 μm) for particles over a narrow range of 0.15 to 20 μm in diameter. Additional measurements and improvements will be made in a flux measurement system that utilizes an integrating nephelometer, to assess the systems' limitations and general usefulness. Signals will be processed primarily by analog signal multipliers but digital methods will also be used for verification. Multiple and simultaneous applications of these sensors will allow comparison for profile and eddy-correlation measurements as well as correlation of the various measurements with pertinent meteorological factors. The data collected in this measurement program will also allow for the direct evaluation of the modified Bowen Ratio and Variance Methods for determining v_d. In addition, a test program will be conducted with other researchers, possibly at Argonne National Laboratory, where simultaneous measurements by various groups of investigators will be compared and evaluated.

Title:

Environmental Contaminants: Effects on Tumor Growth and Immunity

Principal Investigator:
Nancy I. Kerkvliet

Institution:
Oregon State University

Project Period:
2 years

Budget:
(3 years) $179,272

EPA Project Number:
R 808204

Summary:

The primary objective of this proposal is to confirm and extend preliminary findings that exposure of adult mice to pentachlorophenol is immunosuppressive resulting in profound enhancement of both transplanted and virus-induced tumor growth. The three specific aims of the proposed research are: (1) to identify, by use of a battery of *in vitro* immunological assays, the cell type(s) and function(s) altered by PCP exposure which contribute to enhanced tumor growth; (2) to extend these findings to prenatal/neonatal exposure studies, incorporating tests for both host susceptibility to tumor growth and the *in vitro* assessment of immune function; and (3) to use those assays which are most sensitive and predictive of altered immune function in PCP-exposed animals and identify the exposure levels and duration of exposure to PCP necessary to induce immune dysfunction. A complete battery of immunologic assays will be utilized in these studies to determine the cell type(s) and function(s) of the immune system altered by PCP exposure. Both general immunocompetence assays such as mitogen stimulation and mixed lymphocyte reaction as well as assays specific for anti-tumor immunity will be incorporated. The experiments proposed in this project will provide the opportunity to compare the various *in vitro* assays for sensitivity, reproducibility and ability to reflect altered *in vivo* response. A preliminary objective of this research is to establish whether tumor enhancement and immunosuppression are associated with exposure to pure PCP. Animals have been placed on diets containing 500, 1000 or 2000 ppm pure PCP or, as a positive control, 250 ppm technical (86%) PCP. After 12 weeks of exposure, the animals will be injected with Moloney Sarcoma virus. Regressor animals will be inoculated with a progressive dose of MSB sarcoma cells. Tumor growth parameters will be monitored.

Title:

Pulmonary Effects of Ozone and Benzo(a)pyrene in Rats with Altered Antioxidant (Selenium and Vitamin E) Defense Potential

Principal Investigator:
Edward J. Massaro

Institution:
Pennsylvania State University

Project Period:
2 years

Budget:
(2 years) $246,522

EPA Project Number:
R 807746

Summary:

The objectives of this project are to study verified vitamin E and/or selenium deficiencies in the rat and to investigate the effects of the deficiencies on pulmonary lipid metabolism, pulmonary antioxidant defense mechanisms, alveolar macrophage function, pulmonary prostaglandin metabolism, and the function of the mixed function oxidase system of the lung. The following hypothesis will be tested: uncontrolled lipid peroxidation, a potential consequence of inadequate selenium and vitamin E nutrition, can result in the damage of various types of pulmonary cells, subcellular organelles and lung defense functions, including the metabolism of xenobiotics. Both *in vivo* and *in vitro* procedures will be utilized to evaluate aspects of total lung and surface-active lipid metabolism, the lung's susceptibility to membrane lipid peroxidation, its response to potential mutagenic and/or carcinogenic insult resulting from oxidative metabolism of benzo(a)pyrene, and the effects of prostaglandin-like cyclic endoperoxides on the levels of cyclic nucleotides in lung tissue. The research methods to be employed include spectrophotometric and isotope assays of enzyme activity; polarographic determinatoin of oxygen utilization by isolated type II cells and mitochondria; *in vivo* incorporation of tritium from ^3H_2O into pulmonary lipids; isolation of type II alveolar cells; and thin layer and high pressure liquid chromatographic techniques for the analysis of membrane lipids, prostaglandin and benzo(a)pyrene metabolites. Cyclic nucleotide levels will be determined by radioimmunoassay. Phagocytic and bactericidal activity of alveolar macrophages will be assessed by microscopic determination of ingested bacteria and the viability (colony-forming capacity) of a temperature-sensitive mutant of *E. coli*. Antineoplastic activity of macrophages will be determined by isotope procedures involving labeled erythrocytes and lymphocytes and the lysis of several tumor cell lines.

Title:

Case Control Study of Water Quality and Adenomatous Polyps

Principal Investigator:
Jonathan Amsel

Institution:
University of Pennsylvania

Project Period:
18 months

Budget:
(1½ years) $125,220

EPA Project Number:
R 807750

Summary:

This project will explore the possibility of an association between the occurrence of colonic polyps and the quality of drinking water. A case-control design will be used comparing colonic polyp patients diagnosed by double contrast barium enema with nonpolyp patients who have had a double contrast barium enema which showed no abnormalities of any kind. Differences in water quality, based on residential history, between cases and controls will be examined. (Much of this information has already been collected.) Residential history, consumption of drinking water and selected other information will be obtained from the subjects through mailed questionnaires. Telephone followup and, when necessary, personal interviews will be used for individuals who do not respond to the questionnaires. Results of this study will have application with regard to detection of individuals at high risk for polyps. In addition, information about the health impact of constituents in drinking water can be used to set more meaningful standards.

Title:
A Study of Diesel Exhaust Effects in Railroad Workers

Principal Investigator:
Frank E. Speizer

Institution:
Peter Bent Brigham Hospital

Project Period:
2 years

Budget:
(2 years) $973,646

EPA Project Number:
R 807515

Summary:

This study is designed to evaluate the effects of chronic exposure to diesel exhaust. Exposure will be quantified and characterized as to the nature of the polycyclic hydrocarbons. Extensive monitoring will be carried out in selected railroad yards employing two distinct epidemiologic approaches. (1) A group of approximately 80,000 men aged 50 to 64 (in 1964) and having more than 10 years of exposure will be traced to 1980. Total work exposure to diesel exhaust and the outcome, in terms of mortality by specific cause, will be determined. (2) A case control study of incident deaths totalling approximately 300 lung cancer cases, 300 chronic respiratory disease cases and 600 controls in current and retired railroad workers to age 80 will also be conducted. Next-of-kin interviews will be used to determine detailed smoking and residence histories and occupational exposure will be determined from the Railroad Retirement Board's records.

Title:

Economical Solution for Bulking Problem Associated with Activated Sludge

Principal Investigator:
Yeun C. Wu

Institution:
University of
Pittsburgh

Project Period:
2 years

Budget:
(2 years) $103,204

EPA Project Number:
R 807600

Summary:

The proposed research is to investigate the kinetic theory of species selection and to demonstrate the combination of microbial selectors with plug-flow reactors instead of the conventional single reactor plug-flow system. Reactor systems can successfully eliminate the development of filamentous organisms to a certain extent so that the settling properties of the activated sludge will not be significantly affected by crucial factors, such as the organic substrate loading rate. The objective of this study is five-fold: (1) to determine the optimum volume ratio of the selector to the plug-flow reactor under a wide range of substrate and operating conditions; (2) to collect kinetic information from the modified activated sludge system and then to compare the data with that generated from the conventional plug-flow activated sludge system; (3) to study the morphology and structure of activated sludge in response to growth conditions; (4) to determine the most effective alternative to the control of sludge recycle in the modified activated sludge system; and (5) to estimate the cost-effectiveness of the proposed activated sludge system. The experiments will be performed in two 50-gallon activated sludge pilot plants which are constructed of plexiglass. Both reactor systems are to be operated with cellular recycle at room temperature under the controlled pH of 7.0. The quantity of oxygen added to each system will be precisely controlled to insure that the dissolved oxygen content of the mixed liquor suspended solids will be maintained at the desired level throughout the study. Synthetic and domestic wastewaters will be used to feed both activated sludge systems.

Title:
Atmospheric Hydroxyl Experimental Studies

Principal Investigator:
Thomas M. Hard

Institution:
Portland (Oregon)
State University

Project Period:
2 years

Budget:
(2 years) $198,502

EPA Project Number:
R 807733

Summary:

Hydroxyl (HO) is sunlight's chief agent in cleansing the lower atmosphere of trace gases that do not dissolve in rain. In urban air, however, intermediate products of this cleansing process have unwanted effects. This project seeks to obtain an empirical relation between daylight spectral intensities and HO under various urban air compositions, and to achieve better theoretical understanding of the photochemical mechanism that maintains HO. To attain these goals, experiments will be conducted to: (1) improve the accuracy and sensitivity of the determination of atmospheric HO by laser-excited fluorescence; (2) monitor ambient HO and the photochemical quantities that govern it; and (3) observe the response of HO to strong modulation of daylight. Substantial gains in sensitivity have been achieved by expanding the sampled air, folding the laser beam, and in time discrimination. Spectral and temporal characterization of interferences is in progress.

Title:

Trophic State Assessment and Prediction by Aeromonas hydrophila *Densities*

Principal Investigator:
Victor J. Cabelli

Institution:
University of Rhode Island

Project Period:
2 years

Budget:
(2 years) $120,745

EPA Project Number:
R 807252

Summary:

The overall objective of the project is to further explore a previously shown relationship of the *A. hydrophila* densities in fresh bodies of water to their trophic states. It is proposed that the data base be expanded by documenting relationships as evidenced through studies of additional lakes during the summers of 1981 and 1982. Growth studies will be conducted to determine: (1) whether the growth kinetics of *A. hydrophila* seeded into raw and/or treated (heat, filtration, etc.) water samples also correlate with the trophic state of the body of water; (2) nutrients whose concentrations in oligotrophic waters limit the growth of the organisms; and (3) interactions with other limnetic biota (i.e., predation, competition, antibiosis) which also affect the ultimate *A. hydrophila* densities obtained and the observed relationship to the trophic state. Experimental conditions have been defined wherein *A. hydrophila* multiplies in samples of eutrophic water to final densities of 10^5-10^6/ml with generation time approximately every four hours. Some preliminary data obtained suggest that with mesotrophic and oligotrophic waters, the generation times are longer than in eutrophic waters. However, data have been obtained indicating that interactions with other components of the ecosystem (i.e., predation, competition, antibiosis) also influence *A. hydrophila* densities attained in the laboratory and probably those *in situ*. These studies are being continued.

Title:

Improved Scoring of Chemical Transformation of C3H/10T1/2 Cells

Principal Investigator:
Charles Heidelberger

Institution:
University of
Southern California

Project Period:
2 years

Budget:
(2 years) $367,108

EPA Project Number:
R 808309

Summary:

The objective of this project is to improve the scoring of oncogenic transformation of C3H/10T1/2 mouse embryo fibroblasts by chemical carcinogens. This will be accomplished by developing monoclonal antibodies to cell-surface embryonic antigens common to chemically transformed cells and using these antibodies to provide a radioimmunoassay and an indirect fluorescence assay to quantitate transformation. Studies of transformation will be performed using the scanning electron microscope to determine cell-surface morphological changes. These changes will be correlated with the embryonic cell-surface antigens. The use of activated macrophages to kill selectively transformed cells, thus scoring for transformation, will also be explored.

Title:

Retardation of Volatilization Rate of Chemicals by Sediments and Surface Active Layers

Principal Investigator:
R. Thomas Podoll,
James H. Smith

Institution:
SRI International

Project Period:
2 years

Budget:
(2 years) $169,000

EPA Project Number:
R 807710

Summary:

The purpose of this project is to investigate the retardation of the volatilization rate of organic chemicals from water (i.e., retardation caused by adsorption of the organic on suspended sediments and by the diffusional resistance of an insoluble monolayer). To study the effect of adsorption on mass transfer retardation, volatilization rates will be measured in the presence and absence of suspended sediments. Adsorption isotherms will also be measured to determine the amount of chemical adsorbed and to estimate the rate of adsorption. To study mass transfer retardation by insoluble monolayers, volatilization rates will be measured in the presence and absence of insoluble monolayers of long chain aliphatic alcohols. Water evaporation rates will also be measured to normalize monolayer diffusional resistances.

Title:

Anaerobic Degradation of Trihalomethanes

Principal Investigator:
Perry L. McCarty

Institution:
Stanford University

Project Period:
2 years

Budget:
(2 years) $212,712

EPA Project Number:
R 808034

Summary:

Trihalomethanes, produced by chlorination of water supplies, are widely distributed in aquatic environments. Due to the potential health significance of trihalomethanes, their concentration in water is regulated. Previous research has determined trihalomethanes to be readily degradable under anaerobic conditions, while refractory under aerobic conditions. The objective of this study is to further investigate the anaerobic degradation of halogenated organics, particularly trihalomethanes. Laboratory studies will be conducted (1) to determine the biotic and abiotic conditions that lead to the degradation of trihalomethanes; (2) to identify intermediate and end products of degradation through gas chromatographic/mass spectrometric analysis; and (3) to identify factors affecting rate of degradation, including chemical structure and environmental conditions. Research is currently underway to determine mechanisms and intermediates in trihalomethane decomposition.

Title:

Estimation of Environmental Impact on Population Growth Using Stochastic Dynamic Models

Principal Investigator:
Lev R. Ginzburg

Institution:
State University of New York (SUNY)

Project Period:
2 years

Budget:
(2 years) $120,037

EPA Project Number:
R 807885

Summary:

Natural populations live in unpredictable environments. All characteristics of population growth are subject in some degree to random fluctuations induced by the dynamics of environmental parameters. Standard methods for assessing impacts on natural populations are based on the averaged parameters and therefore can be quite misleading. Even with a positive average growth rate there is a finite probability of extinction (or so-called quasi-extinction) which can be significant if the variability of the growth rate is high. The hypothesis of a stationary age distribution, or any kind of determinism, in the models which are typically used leads to a completely distorted picture of what is actually happening and does not aid the decision makers in making intelligent managerial decisions. Therefore, the proposed project will investigate the opportunity of using stochastic models of population growth for formulating reasonable criteria for environmental impact estimates expressed in probabilistic language.

Title:

Inactivation of Single Particles and Aggregates of Rotavirus by Chlorine and Chlorine Dioxide in Water

Principal Investigator:
Roger Floyd

Institution:
Taylor College of Medicine

Project Period:
2 years

Budget:
(2 years) $156,364

EPA Project Number:
R 807990

Summary:

The objectives of this project are to examine the reactions of rotavirus to chlorine and chlorine dioxide and to determine how the aggregation and adsorption of virus particles affect viral inactivation. Rotavirus is a contaminant of drinking water. It is of considerable importance, therefore, to determine the sensitivity of rotavirus to chlorine, the most common water disinfecting agent in use today, and to chlorine dioxide, a potential replacement for chlorine. In water, virus particles may assume various configurations with respect to each other and to debris in the water—they may occur as single particles, as aggregates, or they may be adsorbed to larger particles of debris. Since the amount of work on chlorine inactivation of viruses is not extensive, considerable work will be necessary to explain the basic mechanisms of viral inactivation by chlorine and other agents. Previous work on a related virus has indicated that single particles of virus are inactivated readily by chlorine. However, when virus particles become aggregated, inactivation becomes seriously impaired. The reaction of both single particle and aggregates of rotavirus to chlorine and chlorine dioxide will be examined and compared.

Title:
Neurobiological Effects of Drilling Fluid on an Intact Mollusc, Aplysia

Principal Investigator:
James E. Kanz

Institution:
Texas A & M
University

Project Period:
2 years

Budget:
(2 years) $78,164

EPA Project Number:
R 807137

Summary:

The objective of this investigation is to determine the lethal and nonlethal effects of oil-rig drilling fluid on the nervous system and behavior of an intact, freely-behaving animal, the marine gastropod *Aplysia*. To do this, recording electrodes will be implanted on the nerves of the *Aplysia* that mediate respiration, cardiac activity, kidney efflux rate, gill and siphon reflexive withdrawal, and neuronal pacemaker activity. The neuronal activity related to these functions will be monitored before and after the introduction of various concentrations of drilling fluid. Extracellular neuronal activity of the *Aplysia* will be analyzed by computer neurons contributing to *in vivo* records. This will provide: (1) a more sensitive bioassay of drilling fluid toxicity than behavioral observations alone; (2) a means to correlate short term neuronal changes (induced by drilling fluid) with long term exposure effects; and (3) a means by which a potential toxin's effects on the nervous system can be monitored in intact, unrestrained animals. Already neuronal recording techniques and procedures for computer analysis of activity from neuronal populations in intact animals have been developed. This research will use these important neurobiological advances to assess the impact of drilling fluids on a marine organism.

Title:

The Effect of Herbicides on the Algal Metabolism of Two Agricultural Stream Communities

Principal Investigator:
Robert J. Kosinski

Institution:
Texas A & M University

Project Period:
2 years

Budget:
(2 years) $117,094

EPA Project Number:
R 807978

Summary:

Federal law mandates the protection of nontarget organisms from the effects of pesticides. One of the first nontarget communities that most pesticides will contact will be an agricultural stream. Yet almost nothing is known about pesticide effects on lotic systems or the ability of those systems to adapt to pesticide exposure. This project will evaluate the effects of four herbicides on artificial stream communities that have had both long term and short term opportunity to evolve herbicide resistance. The herbicides will be Aatrex, Treflan, MSMA and Paraquat, and will be tested in that order. The artificial streams will be outdoor, recirculating wooden troughs lined with plastic sheeting and a layer of pebbles. The streams will be allowed to develop periphyton communities derived from water from two Texan sources: the Little Brazos River, which has had much exposure to agricultural runoff and probably has developed a resistant benthic flora; and Minter Spring, which has had a longstanding chemical stability and no pesticide exposure. Half the streams will colonize with the addition of 0.01 ppm of the test herbicide to the water. The ability of the community to evolve resistance from short term, low level exposure will be evaluated. After three weeks of colonization 0.01, 1 and 10 ppm herbicide will be added. There will be four streams with 0.01 ppm prior exposure and four streams without it for each concentration in addition to four prior exposure and four no prior exposure streams reserved as controls. System responses will be followed for three weeks after herbicide addition. Variables monitored at weekly or more frequent intervals during both the colonization and post-addition period will be community phytosynthesis and respiration (by open water oxygen methods), biomass accumulation (by ATP and chlorophyll concentrations), community composition (by slide colonization) and water chemistry. In 1981 Minter Spring-derived communities will be tested. Approximate starting dates for each herbicide will be April 1 (Aatrex), May 15 (Treflan), July 15 (MSMA) and September 15 (Paraquat). Since the order of herbicides was derived from the approximate times of usage in the field, the dates and herbicide order will be the same for Little Brazos-derived communities.

Title:

Relative Uptake, Disposition and Toxic Effects of Inhaled and Ingested Dichloroethylenes

Principal Investigator:
James V. Bruckner

Institution:
University of Texas

Project Period:
2 years

Budget:
(2 years) $275,128

EPA Project Number:
R 808282

Summary:

Information on the toxic potential of many of the organic contaminants of drinking water is quite meager. Data available on one class of contaminants of particular concern, the short-chain aliphatic halocarbons, is limited largely to exposure via the respiratory tract. Thus, one is faced with the uncertainty of the applicability of inhalation data in predicting risks of toxic injury upon consumption of halocarbons in water. The overall objective of the proposed project, therefore, is to establish the scientific basis for the practice of deriving water quality standards for volatile organics by extrapolation of results from inhalation toxicology studies. A major objective is to compare the uptake and disposition of equivalent doses of halocarbons given orally and by inhalation. Three halocarbons will be studied, namely 1,1-dichloroethylene (1,1-DCE), cis-1,1-DCE and trans-1,2-DCE. Their relative absorption and bioavailability from the G.I. and respiratory tracts of rats will initially be characterized. Blood levels of the parent compounds will be monitored at selected intervals after dosing by gas chromatographic analysis. The rapidity and extent of systemic absorption of a range of doses of each halocarbon will be determined under varying exposure conditions. Based upon these findings, the relative uptake and elimination of ingested versus inhaled halocarbons in brain, liver, kidney and adipose tissue of rats and mice will be delineated. Additional groups of animals will be similarly dosed with ^{14}C-labeled halocarbons in order that the relative disposition and binding of metabolites in the liver and kidney may be determined. The subcellular distribution of covalently-bound radiolabel will also be examined in these tissues. Urinary elimination of radioactivity will be monitored as an additional index of halocarbon metabolism and excretion. Finally, the narcotic potency and hepatorenal toxicity of equivalent oral and respiratory doses of 1,1-DCE and 1,2-DCE will be assessed and correlated with levels of the parent compounds and/or bound metabolites in the respective target tissues. Thereby it can be determined whether the disposition and ensuing bioeffects of halocarbons are dependent not only upon the route of exposure, but upon their propensity for metabolism as well.

Title:
Colloidal Metal Carcinogenic and Toxic Action

Principal Investigator:
Max Costa

Institution:
University of Texas

Project Period:
2 years

Budget:
(2 years) $205,873

EPA Project Number:
R 808048

Summary:

Various tissue culture systems will be used to study the mechanisms of particulate metal induced carcinogenesis. A number of particulate metal compounds will be tested for their carcinogenic activities in the cell transformation assay. (It is not possible to completely test the carcinogenic activity of all metal compounds in one study; however, the cell transformation assay will be applied to those metal compounds which are actively phagocytized since this appears to be a primary determinant of a water insoluble metal compound's carcinogenic or toxic action.) Selected metal compounds having different cancer causing activities will be compared for their uptake, sub-cellular localization and site of cellular action during the early stages of neoplastic transformation. Phagocytosis and distribution of the particulate metals will be studied in intact cells by means of light microscopy; video intensification light microscopy (video recordings of living cells); electron microscopy; and elemental mapping with energy dispersive X-ray analysis integrated with transmission electron microscopy. Parallel biochemical studies will focus upon isolating and purifying cellular components (i.e., isolate nuclei and nuclear DNA, RNA and protein) to determine molecular metal binding sites. Metal concentrations in isolated cell fractions will be measured by means of X-ray fluorescence, while crystalline structure of metal compounds in these fractions will be determined by powder X-ray diffraction. The biochemical and intact cell approach will also be applied to study the effect of inhibitors that disrupt metal induced carcinogenesis or that disrupt specific cellular functions (i.e., colchicine, dansylcadaverine, etc.). Mechanism studies will also focus upon correlating whether carcinogenic metals cause DNA repair, or induce mutations in mammalian cells. These and other studies will help determine whether metals are initiators and/or promotors of carcinogenesis.

Title:

Flocculation and Sedimentation of Polydisperse Suspensions in Water Treatment

Principal Investigator:
Desmond Lawler

Institution:
University of Texas

Project Period:
2 years

Budget:
(2 years) $99,250

EPA Project Number:
R 808192

Summary:

The objectives of this proposed project are to study the behavior of particles (changes in the particle size distribution) in polydisperse suspensions undergoing flocculation and sedimentation; and (2) to use the data gathered in experiments and field measurements to test the validity of previously developed mathematical models for flocculation and sedimentation of polydisperse suspensions in water treatment plants. The approach is to conduct a three phase study consisting of (1) batch laboratory experiments; (2) continuous flow laboratory experiments; and (3) field measurements at a water treatment plant. The principal independent variable for all three phases will be the particle size distribution. Other suspension characteristics and operating parameters will be measured. Predictions of the model for particle size distributions will be compared, using several criteria, with measurements from each phase of the study.

Title:
Combined Testing Protocol for Detecting Environmental Mutagenic Agents

Principal Investigator:
Marvin S. Legator

Institution:
University of Texas

Project Period:
2 years

Budget:
(2 years) $369,103

EPA Project Number:
R 807820

Summary:

Mutagenicity testing of chemicals in animals is often done by conducting a battery of tests in separate animals. This project utilizes an integrated version of this approach in which several tests are conducted simultaneously on each animal exposed to an agent. This combined testing protocol includes metaphase analysis of bone marrow lymphocytes, the micronucleus test in polyachromatic bone marrow erythrocytes, the detection of mutagenic substances in urine and the host-mediated assay using microbial indicators. As an adjunct to the combined testing protocol, sperm abnormalities are evaluated in separate animals. The proposed studies will evaluate several chemicals and develop several additional procedures to be evaluated for possible inclusion in the protocol. Chemicals to be tested include acrolein, naphthalene, toluene, and 4,4-methylenediphenyl disocyanote. Additional procedures to be evaluated for possible inclusion in the protocol include detection of DNA damage by alkaline filter elution techniques with bone marrow cells, use of neoplastic transformation and mammalian cell point mutation assays as indicators in the host mediated assay, use of an immunological assay for Hy antigen for detection of mutations and detection of chromosomal abnormalities in germinal cells.

Title:
Molecular Biology, Pathogenicity and Ecology of Bacterial Plasmids—A Conference

Principal Investigator:
Stuart B. Levy

Institution:
Tufts University
School of Medicine

Project Period:
1 week

Budget:
(3 months) $10,125

EPA Project Number:
R 808502

Summary:

Organization, planning and implementation grant for an international meeting on plasmids, Santo Domingo, Dominican Republic, January 5-9, 1981.

Title:
Colo-Rectal Cancer, Water Contaminants and Related Morbidity

Principal Investigator:
Marise S. Gottlieb

Institution:
Tulane University
School of Medicine

Project Period:
2 years

Budget:
(2 years) $367,144

EPA Project Number:
R 807804

Summary:

The purpose of this project is to conduct a case/control study of 250 incident cases of rectal and 300 incident cases of colon cancer. An attempt will be made to determine whether there is a mortality risk of rectal cancer from drinking water. Such a risk has not been noted for colon cancer. Other associated morbidity will also be studied. Four years of incident cases diagnosed within one year of hospital record in eight parishes (counties) of residence at diagnosis will be surveyed. All area hospitals and those hospitals receiving referrals from the area will be surveyed for cases. Controls will be matched to cases by age at death, sex, race, and residence in one of the study parishes. Other disease mortality will be compared to rectal cancer, as well as available morbidity data. Personal interviews will be conducted for cases and controls on lifetime water source, residence, occupation, personal habits (including cigarette smoking), diet, and illnesses. Samples from water sources will be analyzed by gas chromatography and mass spectrometry for possible contaminants from past industrial or other contaminations. A retrospective incidence study for selected confirmed high and low risk communities with identifiable water sources will be designed.

Title:
Developments at the Tenth International Conference on Water Pollution Research

Principal Investigator:
R. S. Engelbrecht

Institution:
United States of America National Committee for the International Association on Water Pollution Research

Project Period:
1 year

Budget:
(1 year) $39,386

EPA Project Number:
R 807269

Summary:

This project provides a report which gives a current assessment of developments in water pollution research throughout the world. The report is based on information gathered at the Tenth International Conference on Water Pollution Research, held at Toronto, Canada, June 23-27, 1980. The Conference brought together research engineers and scientists, consulting engineers, manufacturers of process control equipment, administrators of water quality control programs and others involved in water pollution control. Approximately 30 of the engineers and scientists attending the Conference from the U.S.A. participated in the preparation of this report. Each participant submitted a summary on developments at an assigned session of the Conference for inclusion in the overall report. Both formal and informal developments are covered, including pertinent comments and ideas brought out in discussion of presented papers and among attending researchers. Copies of the report, "Developments at Tenth International Conference on Water Pollution Research," EPA-600/9-81-008, dated November 1980, are available from the EPA.

Title:

Effect of Mean Cell Residence Time on the Thickening and Dewatering Characteristics of Waste Domestic Sludges

Principal Investigator:
W. R. Knocke

Institution:
Virginia Polytechnic
Institute and
State University

Project Period:
2 years

Budget:
(2 years) $59,140

EPA Project Number:
R 807149

Summary:

The objectives of this study are: (1) to determine the effect of mean cell residence time on the thickening and dewatering rates of both biological sludges and primary-biological sludge mixtures; and (2) to evaluate the applicability of utilizing laboratory-scale treatment units to model full-scale operations. The experiments will involve the monitoring of both a laboratory-scale biological treatment reactor and a full-scale (15 MGD) activated sludge treatment plant at several O_c values between 5 and 18 days. At each O_c value, both plants will be monitored for overall treatment efficiency, sludge thickening rates, sludge dewatering rates and solids handling characteristics. The major benefit will be the development of O_c as an operational parameter which can be utilized by waste treatment plant operators to optimize their systems both with respect to treatment efficiency and solids handling.

Title:

Assessment of Impact of Highway Heavy Metals on Roadside Streams

Principal Investigator:
John J. Ney

Institution:
Virginia Polytechnic
Institute and
State University

Project Period:
2 years

Budget:
(2 years) $89,436

EPA Project Number:
R 808138

Summary:

The objectives of this project include: (1) description of cumulative contamination of water, sediments and biotic components of roadside stream ecosystems by highway-generated lead, cadmium, nickel and zinc following inception of highway usage; (2) analysis of changes in abundance, diversity, and distribution of aquatic plants and benthic invertebrates relative to heavy metal contamination; and (3) assessment of reproduction and distribution of fish populations. The approach incorporates temporal and spatial comparisons of metals contamination and population parameters. Two small softwater streams near Richmond, Virginia, will be studied over a three-year period at three sites: (1) upstream, (2) at highway contact, and (3) downstream. The study will focus on a new major highway and will encompass the period prior to highway opening through two years of operation. Since the project was authorized in October 1980, baseline metals and population data have been collected. It is anticipated that the highway will open for automobile traffic in the spring of 1981.

Title:
Prostaglandins as Markers of Promoter—Virus Cell Transformation

Principal Investigator:
Moseley Waite

Institution:
Wake Forest University

Project Period:
2 years

Budget:
(2 years) $134,162

EPA Project Number:
R 807770

Summary:

The purpose of this project is to determine the role of prostanoids in the two-step virus-(co)carcinogen interaction that leads to tumorigenecity, and to establish whether the production of prostanoids in this process is obligatory and therefore, a reasonable marker for (co)carcinogens. Newly-developed clonal lines of HSV-2 transformed tumorigenic or nontumorigenic rat embryo fibroblasts will be utilized. Experiments will involve treating the clonal cell lines with single or multiple doses of the (co)carcinogen phorbol esters. The cells will then be analyzed for alterations in transformation specific (cell morphology, hexose transport, plasminogen activator, phospholipid and fatty acid composition) and tumorigenic specific (efficiency of colony formation in soft agar medium, saturation density and tumorigenicity in newborn adult rat) properties in order to determine the origin and structure of the prostanoids produced under these conditions, and the relationship between their production and the growth characteristics will be studied. A variety of other (co)carcinogens will be tested under conditions expected to produce tumorigenicity. These experiments should assist in understanding a possible two-stage process in malignant transformation by oncogenic viruses.

Title:

Measurement of Tropical Hydroxyl Concentrations

Principal Investigator:
Malcolm J. Campbell
John C. Sheppard

Institution:
Washington State University

Project Period:
1 year

Budget:
(1 year) $104,040

EPA Project Number:
R 807741

Summary:

This project proposes to measure the ambient hydroxyl radical concentration, for periods long enough to secure meaningful averages, at three clean tropical sites. The data, when added to those collected at two Carribbean sites, should make it possible to estimate the latitudinal variation of the hydroxyl concentration in the region where most of the global oxidation of several longer-lived pollutants occurs. This group of pollutants includes the important chlorine-containing solvents, such as methyl chloroform, dichloromethane and others under consideration, which have lifetimes long enough to avoid local ozone production but whose impact on the stratosphere is presently difficult to evaluate. The proposed measurements will complement hydroxyl data being collected at other latitudes and will be accompanied by concurrent measurements of ozone, NO_x, water vapor and ozone-photolytic sunlight flux so that checks may be made of the largely unvalidated models currently dependend upon for prediction of oxidation rates. The experimental approach will be to use a ^{14}C tracer method, developed by Campbell and Sheppard, which involves measuring the rate of $^{14}CO + OH \rightarrow {}^{14}CO_2 + H$ reaction. Because OH is at a photo-stationary state and the slowness of the conversion of ^{14}CO to $^{14}CO_2$, it is possible to obtain hydroxyl radical concentration. A large UV-transparent Teflon bag (100 l) or a quartz reactor are used. Because of the very low conversion of ^{14}CO to $^{14}CO_2$, low-level ^{14}C counting systems must be used to detect the few counts per minute of ^{14}C formed.

Title:

An Atmospheric Tracer Investigation of Flow Around an Isolated Hill

Principal Investigators:
Brian Lamb
Elmer Robinson

Institution:
Washington State University

Project Period:
18 months

Budget:
(2 years) $128,900

EPA Project Number:
R 807336

Summary:

An investigation related to the transport and dispersion of pollutants emitted in complex terrain will be conducted by releasing tracer gases near an isolated hill during periods of neutral and stable stratification. During the tracer experiments, sulfur hexafluoride (SF_6) and bromotrifluoromethane ($CBrF_3$) will be released simultaneously from two different heights upwind of the hill. Measurements of surface and elevated tracer concentrations will be combined with meteorological data and visual analyses of balloon and smoke releases to yield a comprehensive description of flow around an isolated hill. In particular, the tracer data will be used to quantify the conditions where a plume impinges upon a hill and passes round it and also those conditions where a plume passes over the top of a hill. The results of this study will be used to verify results from similar small-scale fluid modeling experiments. The determination of flow patterns in a relatively simple terrain situation will thus provide a basis for extrapolating analyses developed in the laboratory to analyses of flow in complex terrain.

Title:

Dynamics of Polynuclear Hydrocarbons [sic] in an Estuarine Environment Near an Urban Center

Principal Investigator:
Foppe B. DeWalle

Institution:
University of Washington

Project Period:
2 years

Budget:
(2 years) $150,385

EPA Project Number:
R 808337

Summary:

Polycyclic aromatic hydrocarbons (PAHs) occur widely in surface waters and sediment and there is widespread concern regarding the health implication of these potentially carcinogenic compounds. Most PAHs are generated in urban centers from stationary or mobile combustion processes and are redeposited on the ground surface. During rainstorms and subsequent runoff, these compounds can enter the surface water and become associated with the bottom sediments. After deposition in the sediments, the PAHs can be degraded by microorganisms or can accumulate in the food chain; the extent depends on the complexity of the ring structures. The research goal is to provide a better quantitative understanding of the PAH pathways in the estuarine environment near an urban center. The specific objective is to quantify the type of rate of PAH input from various urban runoff discharges and to determine the degree of association between PAH and different types of suspended solids during discharge and after discharge into the estuarine surface water. The separation of soluble and suspended PAHs will be accomplished with a unique depth filtration/resin adsorption system. A similar technique will be used to recover the different PAHs at various distances away from the shore. A vacuum pumping system will be used to collect sufficient quantities from the upper and lower portion of the water column. Sediment traps will also be installed at the sampling station to determine whether the expected decrease of PAHs in the water column corresponds with a parallel increase of the PAH in the settled solids. Tests will be conducted both in the winter and summer when different particulates such as algae and crustaceans are present in the surface layers as they may influence the amount of soluble PAHs. The subsequent research effort will focus on the attenuation of the PAHs by bacterial and following ingestion of sediment particles by benthic organisms. Different types of sediment samples will be evaluated with time to quantify the PAH decrease as influenced by clay content, bacteria concentration and benthic organisms.

Title:

Ecological Impacts Associated with the Use of Fiberglass Screens for Aquatic Plant Management

Principal Investigators:
Michael A. Perkins
Donald R. Baldwin

Institution:
University of Washington

Project Period:
2 years

Budget:
(2 years) $99,474

EPA Project Number:
R 808349

Summary:

The use of fiberglass screens for the control of nuisance growth of aquatic vascular plants has been shown to be highly effective. The ecological impacts associated with the use of these screens would appear to be minimal, however, previous studies have been limited in scope and the extent of ecological impact is in need of additional examination. This current study addresses both water column and benthic effects using a combination of laboratory and field investigations. The hypothesized effects relate to (1) the release of inorganic nutrients and oxygen demanding organics as a result of plant decomposition beneath screens and (2) direct effects upon benthic invertebrate communities. Organic and inorganic nutrient release is being tested in laboratory growth chambers containing Eurasian watermilfoil. Impacts upon benthic invertebrate communities are being evaluated by field application tests. The data generated from these tests should allow for predictive estimation of effects for screen applications under a variety of conditions.

Title:

Measurement of Particulate Nitrogen and Sulfate Compounds

Principal Investigator:
Timothy V. Larson

Institution:
University of Washington

Project Period:
2 years

Budget:
(2 years) $98,898

EPA Project Number:
R 807569

Summary:

The objective of this project is to develop new techniques for *in situ* detection of nitrate and sulfate compounds found in atmospheric aerosol. To achieve this objective, a sampling system that can selectively remove HNO_3 or both HNO_3 and particulate NH_4NO_3 from the aerosol mixture will be designed. This differential sampling system will be used with a nephelometer and a chemiluminescent NO_x detector in order to obtain real-time, *in situ* measurements of NH_4NO_3. In addition, combined temperature and humidity controlled nephelometry for the detection and quantification of H_2SO_4 and its ammonium salts will be explored. These techniques will eliminate several classical interferences such as nitrate salts found in conventional humidity controlled nephelometry and will give a unique signature for compounds ranging from H_2SO_4 to $(NH_4)_2SO_4$.

***Title*:**

Rapid Screening of Chemical Mutagens and In Situ *Monitoring Using Tradescantia-Micronucleus Test*

***Principal Investigator*:**
Te-Hsiu Ma

***Institution*:**
Western Illinois University

***Project Period*:**
2 years

***Budget*:**
(2 years) $96,525

***EPA Project Number*:**
R 807497

Summary:

The major objective of this project is to study the effects of a selected group of chemical agents (including known mutagens, carcinogens, pesticides, food additives and common drugs) by using a rapid test system—Tradescantia-Micronucleus bioassay. This will involve exposing the young inflorescences of Tradescantia plant cuttings to gaseous or liquid forms of chemicals and then observing the micronucleus frequencies in tetrads of the meiotic pollen mother cells. Additionally, the same bioassay will be used to carry out *in situ* monitoring of air at pollution sites—industrial complexes, parking garages, and truck and bus stops. The plant cuttings will be carried to these sites where, after a given duration (6 hours or less), the micronucleus frequencies in tetrads of the meiotic pollen mother cells will be observed. Due to the simplicity and high efficiency of this bioassay, a large number (150) of chemicals and repeated *in situ* monitoring (30) can be accomplished with a limited work force and facility within a relatively short duration of time (3 years).

Title:
Environmental Asbestos and Mesotheliomas

Principal Investigator:
William A. Croft

Institution:
University of Wisconsin

Project Period:
2 years

Budget:
(2 years) $130,991

EPA Project Number:
R 808377

Summary:

Mesothelioma, a neoplasm of the abdomen or chest lining, has been considered a pathological curiosity in domestic animals. In dairy calves, for example, only 16 cases were reported in the 46 years from 1921 to 1967. It is of interest, therefore, that during a two-year period (1977-78) there were 7 histologically-confirmed cases reported among 529,077 calves slaughtered at a single abattoir in northern Wisconsin. This high and unexpected incidence at slaughter has prompted an examination of the geographical and temporal dimensions of mesothelioma in mid-Western dairy herds and its possible relationship to environmental asbestos, the primary etiological agent in the induction of the human disease. Relationships have been established with abattoirs in Wisconsin and Minnesota to provide histological confirmation of all neoplasms identified. Cases will be traced back to their farms of origin and the general farm environment, particularly water supplies, will be examined for possible sources of asbestos exposure. Dairy cattle are readily available as an in-field experimental system and may allow an assessment of the utility of bovine cancers in serving as an index of environmental hazard.

Title:
Predict Reproduction: Toxicants, Pathogens, Climate

Principal Investigator:
Ronald D. Hinsdill

Institution:
University of
Wisconsin

Project Period:
2 years

Budget:
(2 years) $397,773

EPA Project Number:
R 807540

Summary:

The objective of this project is to assemble the following into an integrated model: new heat and mass transfer models for insulated animals, microclimates, outdoor respiratory water loss, physiological thermoregulation, and fractional and full factorial designs. This integrated model will be used to predict the physiological stresses imposed by climate, pathogens, and toxicants and to assess the consequences for reproductive capabilities. Specific objectives include development of a quantitative model that will assess the impact of toxicants, pathogens, and climate on small mammal growth and reproduction in the lab (short term) and in field (long term); identification of physiological parameters that can be used to screen potential toxicants in the laboratory and in the field; identification of physiological parameters that indicate synergistic effects of toxicants, pathogens, and climate; and assessment of mechanisms of action when synergistic effects are found between toxicants, pathogens, and climate. *Peromyscus maniculatus* will be exposed to various environmental toxicants in combination with various types of pathogenic stress. The effect on growth and reproduction will be noted. The model should ultimately provide a more realistic assessment of the effects of environmental contaminants on mammalian systems under real life conditions of exposure. Work done so far includes the breeding of colonies, surveying natural pathogens, completing the computer modeling, and obtaining data from known immunosuppressants.

Title:
Oxygen Uptake Rates for Activated Sludge Process Control

Principal Investigator:
Jerry Y. C. Huang

Institution:
University of Wisconsin

Project Period:
2 years

Budget:
(2 years) $117,285

EPA Project Number:
R 808201

Summary:

The proposed research introduces a new method for expressing the substrate loading rate of an activated sludge process. The proposed substrate loading rate is Fr/Mr, where both terms are derived from oxygen uptake rate measurements and are amenable to rapid and continuous measurement. The research will be conducted initially in a pilot scale activated sludge unit, and subsequently in a full-scale treatment plant. Detailed tasks include: (1) step load increase, plug flow, treating synthetic wastewater; (2) toxic substance, plug flow, synthetic waste; (3) step load increase, complete mix, synthetic waste; (4) toxic substance, complete mix, synthetic waste; (5) step load increase, plug flow, actual waste; (6) toxic substance, plug flow, actual waste; (7) step load increase, complete mix, actual waste; and (8) toxic substance, complete mix, actual waste.

Title:

Oil and Related Contaminant Effects on Waterfowl Immune Defenses

Principal Investigator:
T. M. Yuill

Institution:
University of Wisconsin

Project Period:
2 years

Budget:
(2 years) $124,825

EPA Project Number:
R 808367

Summary:

The objective of the project is to assess the effects of crude oil petroleum distillates and selected chemicals (used to retrieve spilled oil or treat oil-covered birds) on the resistance of waterfowl to bacterial or viral infection. Also, their immune responsiveness (both antibody and cell mediated) will be evaluated. The study will have four phases. First, assays for measuring immunosuppression will be developed for mallard ducks to be used for later assessment of oil and oil-related pollutants. In the second phase, toxicity studies will be conducted to determine mallard sublethal oral doses of the various contaminants. Mallard infectivity and lethality of an avian cholera strain to be used in subsequent experiments will be determined in the third phase. Finally, the hypothesis that ingestion of oil and oil-related contaminants substantially alter immune responses of waterfowl and host resistance to infection will be tested. Currently, toxicity studies have been conducted for one crude oil and one fuel oil. Studies are underway to determine sublethal doses of a dispersant and a dispersant oil combination. Progress has been made on developing additional techniques for measuring immunosuppression. Also, several experiments to determine the infectivity and lethality of avian cholera for mallard ducks have commenced.

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Title:

Atmospheric Aerosol Chemistry of Sulfur and Nitrogen Oxides

Principal Investigators:
L. Robbin Martin,
Henry S. Judeikis,
Donald E. Damschen

Institution:
Aerospace
Corporation

Project Period:
2 years

Budget:
(2 years) $169,150

EPA Project Number:
R 808585

Air pollution by sulfur and nitrogen oxides is a major problem for society, since it is both costly to control and costly to ignore. Public awareness is growing about the problems attendant to this pollution, namely atmospheric aerosols and acid rain. There is considerable urgency about finding cost effective ways to control these problems.

Limiting the emissions of sulfur dioxide has been only sporadically effective in controllilng the sulfate and acidity levels. This may be due in part to changes in emission modes, but also seems to be due to our limited knowledge of the atmospheric chemistry that converts SO_2 to sulfate. Only three years ago, it was not possible to account for the rapidity of formation of sulfuric acid from SO_2 emission, nor was it possible to understand the poor correlation between SO_2 and sulfate levels in the atmosphere.

As a result of recent development in our understanding of the atmospheric chemistry, it is now possible to point to processes that can account for the rapid oxidation of SO_2. These are oxidation by OH radicals in the gas phase, and oxidation by H_2O_2 or ozone in the liquid phase of aerosols. Our preliminary calculations show that the hydrogen peroxide process can easily be the fastest route if any significant amount of aerosol is present. Understanding the correlation problem will require more kinetic data.

Another recent change has been the recognition that nitric acid is a significant contributor to the acidity of rain, particularly in the West. Nitric acid derived from NO_x pollution also accounts for much of the increased acidity in areas of the East in the last few years. Therefore, the chemistry of this new source of acidity needs to be understood as well.

A study will be made of hydrogen peroxide – SO_2 reaction and key NO_x reactions in the kind of detail that will be necessary to predict quantitatively the atmospheric oxidation rate of SO_2 under a variety of meteorological conditions.

The objective of the experimental work is to take the two leading candidates for significant aerosol oxidation pathways, namely oxidation catalyzed by solid particles suspended in water and hydrogen peroxide oxidation, and see how they affect the whole nitrogen oxide – sulfur oxide system. We will also look at reactions between oxidants (H_2O_2+NO_2^-, etc.) and reactions between organics (including amines) and nitrite ion and hydrogen peroxide.

Such reactions will be important in determining the steady state concentrations of the oxidants, and also in the possible formation of toxic products. We will include a temperature dependence study for hydrogen peroxide/sulfur dioxide because of its unique significance in sulfate formation in clouds.

The following systems will be examined:

(1) SO_2; suspended particle catalysis: O_2, O_3, NO_x, H_2O_2 oxidants; temperature and pH.
(2) SO_2; metal ion and organic catalysts; H_2O_2 oxidants; temperature and pH.
(3) NO_x; suspended particle catalysis; O_2, O_2, H_2O_2 oxidants; pH.
(4) NO_x; metal ion and organic catalysts; O_2, O_3, H_2O_2 oxidants; pH.
(5) Amine and other organic reactants; metal ion and particle catalysts; NO_x, H_2O_2 oxidants.

The technique will be fast aqueous kinetics in a stopped-flow apparatus equipped with UV absorption diagnostics.

Title:

Evaluation of Crude Oil Spills on Northern Stream Invertebrate and Periphyton Communities

Principal Investigator:
Vera Alexander

Institution:
University of Alaska

Project Period:
2 years

Budget:
(2 years) $209,367

EPA Project Number:
R 808779

The primary goal of the research is to develop a functional understanding of the response of arctic stream systems to crude oil spills. Specifically, the objectives are:

(1) to examine the response of the invertebrate community to a controlled spill;
(2) to quantify the rate at which oil-soaked rocks and sand/silt are recolonized after exposure to crude oil;
(3) to compare toxicity and behavior modification (drift) of invertebrates in an under-ice and in a summer spill;
(4) to contrast the response of an arctic (North Slope of the Brooks Range) to subarctic stream to oil and to compare their recovery; and
(5) to compare the changes in distribution of fish pre- and post-spill.

Experimental oil spills are to be carried out in a tundra and taiga stream just after the stream surface has become covered with continuous ice. The tundra stream selected is a branch of the Toolik River in the foothills of the Brooks Range (68° 15'N, 149° 21'W). The taiga stream is a channel of Poker-Caribou Creek, 31 miles north of Fairbanks, in the foothills of the White Mountains (65° 15'N, 147° 21'W). Before, during and after each spill, invertebrate drift is monitored, and after five days Hess bottom samples are taken in and above the oil spill area. Rock baskets are placed in oiled and unoiled zones of the stream in order to examine colonization after spring break-up. Periphyton biomass and productivity are measured before and after the spill. Experiments in the laboratory are to examine the toxicity of crude oil trapped under ice in comparison to its toxicity in open water to invertebrates.

Following break-up in the Spring, overwintering success in oiled sections and on oiled rock baskets will be examined. Overwinter oil loss from artificial substrates will be determined. In the Autumn of 1982 post-recruitment sampling for the status of recovery one year after the spill will be carried out. Attached algal biomass and distribution will be examined, as will the use of stream sections by fish. Following the sorting and identification of collected invertebrate and algal samples, the data will be analyzed, compiled and reported.

Title:

An Ecosystem Approach to the Effects of Placer Mining on Streams of Interior Alaska

Principal Investigator:
Jacqueline
LaPerriere

Institution:
University of Alaska

Project Period:
2 years

Budget:
(2 years) $214,336

EPA Project Number:
R 808340

Placer gold deposits lie within gravels near bedrock where they occur. Mining for these deposits entails removal of overburden, and separation of the gold from the other minerals. Water, under high pressure, is sometimes used to strip overburden, and to thaw any ice it contains in permafrost regions, in an operation called hydraulicking. Water is also used in the separation process called sluicing. The water used in placer mining often comes from and is discharged into nearby streams.

Even when sedimentation ponds are used to treat the effluent from placer mining operations, there is a potential for the addition of sediments in amounts or at times that are not natural. This is especially true where the soils or gravels contain clays, or when sedimentation ponds fail due to flooding, short-circuiting, or breaching of their walls.

The effects of additional sediments from placer mining on the hydrology, sediment carrying capacity, and organisms of the aquatic ecosystem of streams are not well known. This project will investigate the effects of sediment and bedload-transport on periphyton, on benthic macroinvertebrates, and on resident fishes.

Because this industry is governed by the Alaska Water Quality Standards for turbidity, total dissolved solids, and sediment, this project relates measured effects on communities or populations in the aquatic ecosystem to specific thresholds for these factors.

Title:

A Solute Model for Predicting Transport of Metals from Industrial Solid Wastes Through Soils

Principal Investigator:
Wallace H. Fuller

Institution:
University of Arizona

Project Period:
3 years

Budget:
(2 years) $162,691

EPA Project Number:
R 807915

Since the land is considered as the ultimate receptacle for industrial wastes the ability to predict attenuation of pollutants in soil becomes one of the main goals in waste control operations. Before predictive designs for disposal of pollutant-containing industrial waste streams can be established, a multitude of disposal components must be identified and evaluated. The three most critical components of the intimate disposal environment affecting transport of a pollutant from its point of origin are: (a) the transport system or fluid carrying the pollutant (leachate and/or solvent); (b) the porous medium through which the constituents move (soil, soil material); and (c) the potential pollutant(s) itself. No two pollutants migrate identically to the same set of soil characteristics of transport fluid properties.

The objective of this research is to quantitatively measure the effects of soil and transport fluid properties on rate of movement of selected metals and certain organic pollutants to aid in the development of a solute model for predicting the transport of pollutants from industrial and commercial wastes through soils. The effects of the waste stream transport systems such as (a) dilute aqueous fluids, (b) strong acids, (c) strong bases, (d) polar organic solvents, and (e) non-polar organic solvents on soil properties influencing fluid flux and soil attenuation of pollutants are being researched. A solute model is being developed that will describe the metal transport phenomena discovered in the above research.

The effects of 2*N* acetic, hydrochloric, phosphoric, and sulfuric acids and two sulfuric waste streams (industrial) containing toxic metals on fluid flow through soils are being studied. Data collections on soil failure are being examined and plotted by comparing chemical characterizations of soil column effluents with influents.

The permeability of eight soils to organic solvents and water was investigated. Saturated hydraulic conductivity and intrinsic permeabilities were evaluated. The solvents were kerosine, isopropyl alcohol, ethylene glycol, xylene, and water. Two hundred and eleven column tests were made to evaluate differences between intrinsic permeability for the different soils and solvents. A multiple regression analysis provided a predictive model for intrinsic permeability as a function of soil properties with an R^2-value of 0.81. Moisture retention curves were determined using a pressure retention procedure for three soils and two solvent combinations—water and kerosine. Kerosine results do not follow a general pattern, demonstrating changes in procedure will sometimes become necessary when working with organic solvents in soils.

Title:

Quantitative Measurement of Deposition of Diesel Exhaust Particulates in the Human Respiratory Tract

Principal Investigator:
F. Charles Hiller

Institution:
University of Arkansas

Project Period:
2 years

Budget:
(2 years) $189,710

EPA Project Number:
R 809098

Many atmospheric aerosol particulates and also particulates in the working place are in the size range of less than 0.1 m. There is much theoretical information based on mathematical models of the lungs, suggesting that deposition fraction of particulates less than 0.1 m in the human respiratory tract is quite high. Experimental studies verifying the deposition of such particulates are few.

Most studies describing the deposition fraction of inhaled aerosols are concerned primarily with particles in the 0.1-10.0 m size range. Although the mass of particles less than 0.1 m diameter is quite low, their ubiquitous presence, their predicted high deposition fraction, and their potential for acting as condensation nuclei or carriers for toxic gases could make them important in human disease.

One example of such a particulate is the particulate fraction of diesel exhaust. These particles are mainly less than 0.1 m and carry some carcinogens. Particles in the 0.1 m size range are most difficult to generate and measure. For this reason, experimental studies to characterize deposition fractions in the lung are few. There is need, therefore, for experimental data to confirm or disprove the high deposition of such small particulates.

The purpose of this work is to generate a well-characterized aerosol in the 0.0-0.1 m size range and to measure the deposition fraction of that aerosol in the human respiratory tract.

Title:

Muscle Protein Metabolism After Insecticide Exposure

Principal Investigators:
Edward G. Ezrailson,
Allan J. Garber

Institution:
Baylor College of Medicine

Project Period:
2 years

Budget:
(2 years) $140,936

EPA Project Number:
R 809164

Humans are exposed chronically to varying levels of insecticide as the result of vastly different circumstances and conditions. The greatest degree of exposure is occupationally related. In the manufacture, distribution and harvesting of agricultural products, acute and chronic insecticide exposure may have a direct relationship to the *health and safety* of the participating worker and those in the immediate environment. Safe levels for acute (LD$_{50}$) and chronic (90LD$_{50}$) exposure skeletal muscle contains a metabolically active pool of protein which is in a continual state of turnover, although rates of synthesis and degradation may vary greatly for individual muscle proteins.

Regulation of muscle protein degradation and the subsequent amino acid release is in part under the control of the *autonomic* nervous system. Adrenergic agonists inhibit and cholinergic agonists stimulate protein degradation and the resultant release of amino acids from muscle. Thus, inhibition of acetyl cholinesterase both on an acute as well as a chronic basis may be expected to have profound effects on skeletal muscle protein degradation and homeostasis.

This research will examine the effects of acute and chronic exposure to two model insecticides which have cholinesterase inhibition activity, namely malathion and parathion. Effects of these agents on muscle protein, amino acid and cyclic nucleotide metabolism will be demonstrated. The results expected from this project may be extrapolated to chronic but sublethal clinical problems observed in people exposed to insecticides on an acute and chronic basis, particularly chemical workers, crop dusters, farmers and agricultural workers.

Title:
Transfer of Xenobiotics in Blood to Cells

Principal Investigator:
Louis C. Smith

Institution:
Baylor College of Medicine

Project Period:
3 years

Budget:
(2 years) $216,958

EPA Project Number:
R 808773

The general objective is to determine if polynuclear aromatic hydrocarbons (PAH) entry into cells is controlled by thermodynamics or by the kinetics of lipoprotein modification or by endocytosis. Specific objectives are to define principal mechanisms of cellular uptake of carcinogens in plasma lipoproteins. There may be (a) passive transfer of PAH from plasma lipoproteins to cultured endothelial cells, (b) lipoprotein lipase dependent transfer of PAH in triglyceride-rich lipoproteins to endothelial cells, and (c) cellular uptake of lipoproteins containing PAH.

The experimental methods are based primarily on fluorescence techniques. The concentration dependent fluorescence of pyrene and benzo(a)pyrene allows rapid and continuous determination of changes in PAH mass, as the result of transfer from one membrane to another, without the need to isolate physically reactants and products. Changes in the quenching of the fluorescence of non-transferable carbazol-containing compounds by the polyhalogenated compounds as the PCB are transferred will allow assessment of the kinetics and distribution of the PCB in these systems. The highly sensitive video intensification microscopy will permit direct studies of PAH transfer from plasma lipoproteins to cell membranes and of intracellular PAH transfer from endocytotic vesicles.

The initial step of chemical carcinogenesis is delivery of the carcinogen to the target tissue. In plasma, non-polar substances such as PAH are confined to plasma lipoproteins. Transport of PAH has a regulatory role as a rate determining step. Evaluation of the mechanism of transfer, of the rates of competitive processes, and the thermodynamics of PAH distribution is necessary to understand the relationship of carcinogenesis and lipid metabolism.

Title:
Mechanisms of Human Rotavirus Inactivation by Water Disinfectants

Principal Investigators:
James M. Vaughn,
Edward F. Landry

Institution:
Brookhaven National Laboratory

Project Period:
2 years

Budget:
(2 years) $221,432

EPA Project Number:
809489

The primary objective of the proposed research will be the elucidation of the specific mechanisms of human and animal rotavirus inactivation which result from the use of chemicals commonly employed for the disinfection of potable water supplies. A secondary consideration will involve a comparison of the relative effectiveness of a variety of compounds (e.g., O_3, Cl_2, ClO_2) in the inactivation of rotaviruses.

The proposed research represents the first known scientific investigation of the effectiveness and modes (i.e., specific sites) of rotavirus inactivation by drinking water disinfectants. Data resulting from the study should provide regulatory agencies with an adequate data base with which to assess the reliability of current, or proposed methods for the control of rotavirus species in drinking water. In addition, comparative studies will provide rotavirus inactivation data which is needed for the identification of viable alternatives to the use of chlorine as a water disinfectant.

The objectives of the study will be pursued in the form of specific research goals (presented here with appropriate methods):

(1) A determination of disinfectant thresholds and rates of inactivation of culturable animal rotaviruses (Neonatal Calf Diarrhea Virus [NCDV], Simiam Rotavirus [SA-11]). Viruses will be exposed to various disinfectant concentrations at several pH and temperature levels. Resulting viability data will be analyzed to determine the kinetics of inactivation. Survival rates in various disinfectants will be compared.

(2) A determination of whether gross morphological changes occur on rotavirus particles following disinfectant treatment. Experiments will involve treatments of both human and animal rotaviruses. Analyses will include examination by electron microscopy, as well as determination of particle density via sedimentation velocity measurements of radiolabeled virus.

(3) A determination of the specific site(s) of inactivation on culturable rotavirus as caused by selected disinfectants. Test organisms will include animal (NCDV, SA-11) as well as human rotavirus species (type 2, Wa strain). Investigations will focus on alterations of radiolabeled viral coat proteins (inner and outer capsids) as well as changes produced on labeled segmented RNA. Effects on both will be measured by comparison to untreated controls via polyacrylamide gel electrophoresis. Following initial studies with culturable viruses, similar experiments will focus on non-culturable human rotaviruses isolated from clinical samples.

(4) An assessment of the relative effectiveness of various disinfectants for the inactivation of rotaviruses based upon the results of the above experimental procedures. Analyses of the above data will also provide insight concerning the reliability of animal rotavirus or surrogates for human rotavirus in disinfections studies.

Title:

Test of a Highly Sensitive Drosophila Assay for Chemical Induced Chromosome Breakage

Principal Investigator:
Stanley Zimmering

Institution:
Brown University

Project Period:
3 years

Budget:
(2 years) $182,646

EPA Project Number:
R 808648

Drosophila is the most rapid and efficient *in vivo* complex eukaryote short-term test system for the detection of genetic alterations induced in germ cells by chemical agents. It possesses an efficient microsomal activating system and detects genetic damage of relevance to humans including gene mutations and chromosome breakage; the test for sex-linked recessive lethals assays for gene mutation and tests for chromosome loss and/or heritable translocations for chromosome breakage. Consistent with findings in many other test systems monitoring for two (or more) genetic endpoints, the sensitivity of the detection capacity of Drosophila tests for gene mutation and chromosome breakage is not the same. With the exception of a few compounds, chromosome breakage effects require substantially higher concentrations for detection than recessive lethals or fail detection entirely even at concentrations producing high frequencies of recessive lethals.

Zimmering and co-workers recently reported results from a test for chromosome loss with radically improved sensitivity, achieved by substituting the ordinary repair-proficient Pl females with females homozygous for the repair deficient strain *st mus302*.

The *st mus302* test for chromosome loss has been evaluated thus far with four compounds: methyl methanesulfonate (MMS); dimethylnitrosamine (DMN); diethylnitrosamine (DEN); and procarbazine. Compared with ordinary females (1) *st mus302* females yield significantly higher frequencies of induced chromosome loss with MMS and DMN producing a positive effect at a concentration one order of magnitude below that producing no effect with ordinary females; and (2) more critically, detects chromosome breakage activity with DEN and procarbazine, compounds proved negative in tests with ordinary females at *all* concentrations.

The principal objectives of the proposed research are:

(1) to extend the *st mus302* test for chromosome loss to cover a large number of compounds in diverse chemical groups, comparing its sensitivity relative to the conventional test for chromosome loss, and its sensitivity and overall technical facility relative to the test for heritable translocations in order to determine its suitability as a replacement for these procedures, *in use now for some 20 years*, as a primary screen for chemical induced chromosome breakage in germ cells of Drosophila; and

(2) to carry out experiments to provide the basis for an assessment of the proposal by Sobels and co-workers that the relative insensitivity of breakage tests in Drosophila is attributable to the largely ineffective ability of chemical mutagens to induce *chromosome breakage relative to recessive lethals, data thus far with st mus302* suggesting the alternative view, that relative insensitivity may be less related to *induction* than *detection* capability of the test system.

Finally, demonstration that the *st mus302* test for chromosome loss is no less sensitive than the heritable translocation test, the latter requiring an F2 and often an F3 generation for completion would imply that it be possible to screen a considerably greater number of compounds/time and result in very substantial savings in government sponsored mutagenicity testing programs.

Title:
Submicron Aerosol Classification Using High Velocity, Low Pressure Impactors

Principal Investigator:
R. Flagan

Institution:
California Institute of Technology

Project Period:
3 years

Budget:
(2 years) $107,422

EPA Project Number:
R 809191

The primary objective of this proposal is to study the influence of gas compressibility and Mach number on the performance of aerosol impactors. The first phase of the work will involve theoretical and experimental studies of gas flow and particle impaction in a single stage impactor operating over a wide range of Mach numbers and particle Knudsen numbers. The gas flow in the impactor will be calculated theoretically in the subsonic and transonic regimes using the Lax-Wendroff and other appropriate techniques. The particle trajectories will then be determined, from which the critical Stokes number for particle impaction and the influence of Mach, Knudsen numbers, etc., on the critical Stokes number will be determined. For the experimental investigation, a model single stage impactor will be constructed, and the flow field within the impactor studied by flow-visualization (shadowgraph, Schlieren, etc.). Particle impaction studied will be made by means of monodisperse aerosol generated by the Berglund-Liu monodisperse aerosol generator and the TSI electrostatic classifier using fluorescent or optically absorbing particles.

In addition to the primary research objective indicated above, the study will involve the use of fiber collection surfaces for the impactor and the determination of the effect of fiber collection surface on the impactor characteristics. Also to be investigated are the jet-cone impactor concept developed by Schott and Ranz, and the effect of vapor condensation on impactor operation. Finally, an impactor will be designed, constructed and tested for particle collection from high temperature gas streams.

Title:
A Kinetic Study of the Catalytic Autoxidation of Aquatic Pollutants by Hybrid Metal Complexes

Principal Investigator:
Michael R. Hoffman

Institution:
California Institute of Technology

Project Period:
2 years

Budget:
(2 years) $207,047

EPA Project Number:
R 809198

The kinetics and mechanisms of the oxidation of selected hazardous pollutants by molecular oxygen catalyzed by homogeneous and heterogeneous complexes of cobalt(II) with sulfonated porphyrin, phthalocyanine and polyamine ligands have been investigated. Modified homogeneous catalysts and solid supported analogs have been synthesized and their ability to activate dioxygen with concomitant oxidation of selected reduced sulfur, substituted kinetically. Hybrid catalytic systems were developed by chemically and physically linking effective Co(II) complexes to the following inert surfaces: polystyrene, silica gel, glassy activated carbon, polyacrylamide and zeolite. The catalytic reaction rates, reaction stoichiometries and rate laws were established for both homogeneous and heterogeneous systems in batch and continuous flow reactor systems. Experimental techniques employed in kinetic studies included conventional UV-VIS spectrophotometry, specific ion potentiometry, ion chromatography, high performance liquid chromatography, gas/liquid chromatography, stopped-flow/temperature jump relaxation kinetics. Electron Spin Resonance, Fourier Transform IR spectroscopy, and Resonance Raman spectroscopy were used to determine the modes of dioxygen activation and the relative contribution of radical enzyme-like and polar reaction pathways to the overall aqueous phase oxidation rates of reduced sulfur compounds such as hydrogen sulfide, sulfur dioxide, mercaptans and substituted monosulfides, and substituted aromatic compounds such as 3,4-dihydroxybenzoic acid, 1,2,4-trimethylbenzene and benzaldehyde.

Catalytic activity was determined to be a function of primary liquid and solid phase properties such as pH, temperature, ionic strength, surface area, porosity, degree of cross-linking and the nature of the substrate. Factors affecting the catalyst specificity, lifetime, and modes of inactivation were established. Suitable kinetic and mechanistic models will be discussed. This work is expected to have application in the development of hybrid catalytic systems which may provide convenient and economical methods for industrial wastewater treatment in which the control of oxidizable, hazardous chemicals and refractory organics is desirable.

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Title:

Development of a Valid In Vitro *Assay for Fibrogenesis Using Cultured Pulmonary Macrophages and Fibroblasts*

Principal Investigator:
Stephen C. Benson

Institution:
California State
University-Hayward

Project Period:
3 years

Budget:
(1 year) $104,987

EPA Project Number:
R 808910

Mammalian lung tissue exposed to air pollutant particles and noxious gases undergoes a variety of responses including fibrosis. Most notably this results in increased fibroblast proliferation and collagen synthesis. At present the fibrotic potential of particulate and gaseous material is determined by time consuming *in vivo* fibrogenesis studies. It would be extremely useful to have a reliable *in vitro* model that could be rapidly applied to assess the fibrogenicity of small amounts of material collected from ambient air samples or from combustion processes. The problem with performing extensive tests using *in vitro* model experiments is that the gap between the *in vivo* effect and the *in vitro* model has not been bridged adequately. *In vitro* effects are often found to be artifacts and do not express true mechanisms which operated *in vivo*.

This study will induce *in vivo* fibrogenesis with silica and asbestos and simultaneously carry out *in vitro* studies using cultured alveolar macrophages from the challenged animals. Recently, it has been reported that damaged alveolar macrophages release a fibrosis-stimulating factor (FSF). A reasonable hypothesis is that *in vivo* silica and asbestos-induced injury to alveolar macrophages cause the release of FSF which triggers the increased proliferation of lung fibroblasts and increased collagen synthesis in these cells. The experimental protocol of this proposal will test the hypothesis that an *in vitro* fibrogenic system can accurately mimic the *in vivo* situation and that a totally *in vitro* assay system is feasible.

The successful development of a reliable *in vitro* system to assess the fibrogenic potential of environmental pollutants could potentially obviate the need for large scale long-term animal studies and still provide just as meaningful information. In addition, the diversity in biochemical and morphological responses to asbestos and silica would suggest that these two agents may induce fibrogenesis by different mechanisms. Nevertheless, there is extensive evidence to suggest that macrophages may be directly involved in the fibrogenic response. The proposed research will establish the *in vivo* and *in vitro* relationship between exposure to these agents and the fibrogenic response which ensues. This study will test a reasonable hypothesis regarding how fibrogenesis is induced via biochemical and structural damage to the macrophage, releasing FSF. This information will be vital in establishing a reliable *in vitro* model for fibrogenesis.

Title:

Methods for Assessing the Impact of Environmental Chemicals on Human Fertility

Principal Investigator:
J. Overstreet

Institution:
University of California-Davis

Project Period:
3 years

Budget:
(2 years) $267,565

EPA Project Number:
R 809089

The purpose of this project is to develop sensitive tests for early detection of reproductive toxicity in men and women. Changes in sperm morphology may reflect acute stress to the testes. Morphometric measurements have been made on spermatozoa from 30 fertile men and 30 infertile men. From analysis of these data we have concluded that the degree of abnormality in sperm morphology can be objectively recorded by calculating the ratio of head length to head width and the standard deviation for this parameter. A semi-automatic, computer-assisted methodology has been developed to facilitate these measurements. Studies are in progress to establish a "normal range" for this parameter. The method is now useful for comparison of exposed and control populations.

For tests on living sperm, methods have been developed for transportation of semen from the field to specialized research laboratories. Semen samples from 10 fertile men and 17 infertile men were diluted with an egg yolk extender and stored at 4°C for 48 hours (a temperature and time interval which are feasible for commercially available transportation services). The sperm motility and fertility (as assessed by penetration of zona-free hamster eggs) were preserved in normal and abnormal semen.

Tests of female reproductive toxicity involve detection of early pregnancy and prediction of spontaneous abortion. We have assayed hCG in daily urine samples from more than 100 women during 250 cycles of therapeutic artificial insemination. Fifty of these women conceived pregnancies, all of which were monitored with serial urines during the first trimester. Nine women had spontaneous abortions while in the study. Among the women with no clinical signs of pregnancy a small sub-group had peri-implantation spontaneous abortion (occult pregnancy). The results obtained so far suggest that the incidence of occult pregnancy in normal women may be quite low. The methods we are developing could therefore be useful in surveillance of non-contracepting women who are exposed to potential environmental hazards. We are currently evaluating the possibility of measuring urinary steroid conjugates as an adjunct in detecting early pregnancy wastage.

Title:
Inhalation Toxicology of Formaldehyde-Containing Air Pollutants

Principal Investigators:
T. Timothy Crocker,
Peter Reischl

Institution:
University of California-Irvine

Project Period:
3 years

Budget:
(2 years) $378,873

EPA Project Number:
R 808267

This project will evaluate short-term toxic and irritant effects of combined photochemical pollutants including formaldehyde, particulates and ozone by exposure of rats and dogs during exercise.

The study has the following elements:

(1) Atmospheres: The proposed list of atmospheres for study may be modified by relevant new findings seen in the course of study.
  a. Formaldehyde alone at 0.1 to 5.0 ppm
  b. Formaldehyde plus ammonium sulfate aerosol at 1-4 mg/m^3, equilibrated for gas to particle absorption
  c. Ozone alone at 0.2 to 0.8 ppm
  d. Ozone combined with ammonium sulfate aerosol within the above concentration ranges
  e. Formaldehyde plus ozone plus ammonium sulfate at physical equilibrium with respect to gas absorption in particles
  f. Relative humidity: selected on the basis of biologic endpoint within the range of 40% to 90%
  g. Exposure times: rats 4 hours per day for up to 5 days; dogs 2-4 hours per day for 1, 2, or 3 days.

(2) Exercise: Rats trained to 0.4 m.p.h., at level grade, dogs trained to 3 m.p.h. at level and 15% grade.
(3) Histopathology, quantitative morphometry and indicators of cell turnover for nasal and tracheobronchial epithelia in rats.
(4) Pulmonary function and related measurements in dogs:
  a. Lung dynamics: rate, tidal volume, FRC, airway resistance and compliance, arterial-alveolar oxygen and CO_2 gradients
  b. Metabolic measurements: oxygen consumption and CO_2 production
  c. Breathing pattern with analysis of duration of inspiratory and expiratory phases, pause between phases, air flow rates throughout each phase, and expiratory flow rates at 25% and 50% of tidal volume.
(5) Clearance of insoluble radiolabeled tracer particles from lungs of rats: short-term (mucociliary phase), 0-2 days; long-term (macrophage transport phase), 2-14 days.

Hypotheses to be Tested

(1) Do atmospheres of formaldehyde, formaldehyde combined with sulfate aerosol or formaldehyde plus aerosol plus ozone at concentrations found to be irritant in short-term tests cause histopathologic and functional changes compatible with the potential for producing chronic respiratory tract disease?
(2) What is the quantitative relationship between exercise-enhanced ventilation and the pathogenetic potential of the above single or combined gas-particle atmospheres?
(3) Is the effect of exercise explained solely by the increased minute ventilation or do other as yet unidentified factors play a significant role?

Title:

Cohort Studies of Health Effects of Chronic Exposure to Oxidants and Other Pollutants in Los Angeles

Principal Investigator:
Roger Detels

Institution:
University of California-Los Angeles

Project Period:
3 years

Budget:
(2 years) $228,652

EPA Project Number:
R 808620

This research will continue ongoing studies of the relationship between long-term exposure to air pollutants and the results of a respiratory questionnaire, spirometry, single-breath nitrogen testing and plethysmography in 15,164 residents of four communities in Los Angeles. The study sites include areas exposed to: (1) low levels of all pollutants (Lancaster); (2) moderate levels of photochemical oxidants and other secondary pollutants (Burbank); (3) high levels of photochemical oxidants, NO_2 and sulfates (Glendora); and (4) high levels of SO_2 particulates and other primary pollutants (Long Beach).

Comparison of lung function test results from the baseline studies suggest that participants in the polluted areas, particularly those exposed to high levels of photochemical oxidants, NO_2 and sulfates had poorer respiratory function. Retesting of participants in the two less polluted areas to detect changes in lung function over the last five years has been completed.

This research project will complete retesting of participants in the two remaining areas (exposed to high levels of primary and secondary pollutants), to compare changes in lung function of those in the previously completed areas, and to correlate these with pollutant exposures occurring in the four areas.

Title:

Development and Calibration of the Low Pressure Impactor for High Mass Loading and High Altitude Measurements

Principal Investigator:
S. K. Friedlander

Institution:
University of California-Los Angeles

Project Period:
18 months

Budget:
(18 months) $59,790

EPA Project Number:
R 808786

Cascade impactors are commonly used for size segregation of aerosols for subsequent chemical or elemental analysis. Although a well-established technique, experimental difficulties still exist for the collection of solid or "bouncy" particles. Currently, greased coatings are used on the collection surfaces to prevent particle bounce and reentrainment. However, the coatings become ineffective as the aerosol deposit builds on the collection stage. Laboratory studies have shown that the collection efficiency of a greased impaction surface decreases exponentially with particle loading. The particle bounce problem reappears as the greased surface becomes coated with the aerosol.

This effect is particularly acute for impactors with small cutoff diameters since the mass of very small particles equivalent to the monolayer coating on the stage is less due to the large specific surface area. A low pressure impactor developed in this laboratory segregates particles in the size range from 0.05 to 4 m aerodynamic diameter. It has been successfully used for aerosol sulfur analysis. However, the aerosol coating on the sticky surface severely limits the mass of sample which can be collected. Currently, the instrument cannot be used for reliable distributions of the minor constituents of the aerosol. Submicron elemental size distributions of these trace species would be valuable both for source resolution studies and for understanding the mechanisms of pollutant formation.

The objective of this research project is to develop an impaction surface which (1) has a high collection efficiency for solid (bouncy) particles independent of mass loading and which (2) is suitable for elemental analysis. Specific application will be to the low pressure impactor but the results will be applicable to other samplers. The work will include the recalibration of the impactor with the newly developed impaction surface and recalibration at reduced inlet pressures for high altitude sampling.

Title:
Scattering by Curved Phase-Front Beam (An In-Situ *Scheme to Detect Fine Pollutants)*

Principal Investigator:
Cavour Yeh

Institution:
University of California-Los Angeles

Project Period:
2 years

Budget:
(1 year) $64,140

EPA Project Number:
R 809099

Health hazards caused by the presence of asbestos particles in the atmosphere or in water are well recognized. However, it is not a simple feat to detect and identify such asbestos particles, especially when these particles are of submicron sizes. One way is to make use of the distinguishing light scattering characteristics of a needle-shaped asbestos particle to separate it from other harmless roundish particles. The usual optical scattering technique may be used to obtain substantial amounts of information when the scattering objects are known to be nearly identical (monodisperse) and spherically symmetric. But asbestos particles in the atmosphere are neither spherical (they are needle-shaped) nor monodisperse (there are other kinds of particles in the atmosphere). Hence, it is essential that in some way the scattering must originate from a single isolated particle at a time. A brute force method for accomplishing this isolation involves electrostatically supporting an individual particle while it is illuminated by a laser beam. Another method is to force suspended particles to flow through a micron-size tube which is illuminated transversely by a laser beam. The resulting flashes of light provide information in individual particles. However, in these schemes the environment of the scatterer is somewhat artificial. This fact may be of great consequence when one wishes to obtain more precise information on pollutants, since the mere fact of isolation or being forced to flow through intra-small passageway is likely to alter the properties of the different species in a polydisperse medium. A new nondestructive scheme should be found.

The purpose of this research is to study a new nondestructive diagnostic technique based on the concept of light scattering by a strongly focused laser beam, to identify submicroscopic particles. Initial experiment shows that it is possible to study the scattering characteristics of an individual submicron particle in surroundings approaching its natural environment and in a polydisperse medium. When perfected, this scheme may provide a simple and economical means of measuring the *in situ* concentration and sizes of different species of particles. To reach this goal a two-year program is being undertaken. The program consists of two major tasks:

(1) To obtain scattering signatures of three general types of particles (roundish, long-skinny and disk-like particles) in strongly focused beams, and
(2) To construct an apparatus to measure the scattering characteristics of different particles in a strongly focused beam.

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Title:

The Interrelated Effects of Organic Loading, Dissolved Oxygen Concentration and Aeration Basin Configuration on Activated Sludge Settling Characteristics

Principal Investigator:
David Jenkins

Institution:
University of California-Richmond

Project Period:
2 years

Budget:
(2 years) $132,670

EPA Project Number:
R 808303

The general objective of this research is to further the knowledge in the area of activated sludge bulking at low organic loading rates. Specifically, the objectives will be to examine the effects of aeration basin configuration, aeration basin dissolved oxygen concentration and organic loading on activated sludge settling properties.

Previous work by the Principal Investigator has shown that at low organic loadings increasing the degree of aeration basin compartmentalization generally produces better settling activated sludge. The necessary degree of compartmentalization to consistently achieve such improvements will be determined. In addition, the mechanism by which the installation of small initial aeration basin compartments improves activated sludge settling will be determined.

These studies are being conducted in continuous flow laboratory-scale activated sludge units operating on Richmond, California settled domestic sewage. In these activated sludge units it is possible to control flow and concentration of influent sewage in the aeration basin. In addition to routine performance and activated sludge settling characteristic measurements, a determination of filamentous organism type and abundance in the activated sludge is made routinely.

The mechanisms of activated sludge settleability improvement through aeration basin compartmentalization will involve isolation and physiological/metabolic characterization of the filamentous organisms causing deterioration of sludge settling.

Our current data and previous literature suggests that one possible reason that small initial compartments ameliorate bulking is that in such compartments the activated sludge is exposed to high organic matter levels and storage of these organic substrates takes place. The hypothesis is that filamentous organisms causing bulking cannot store substrates as effectively as floc forming organisms. Thus, conditions favoring substrate storage disfavor filamentous organism growth and lead to well-settling activated sludge. The hypothesis will be tested both in the laboratory activated sludge systems using domestic wastewaters and using the pure cultures of causative filamentous organisms isolated from the activated sludge.

The formation of storage products such as glycogen, polyhydroxybutyrate, and polysaccharide will be investigated. Attention will also be given to the important mechanisms taking place in the large completely mixed aeration basin following the small initial compartments. The necessary detention time, dissolved oxygen levels, etc., will be determined. The utilization of storage products in this basin will be studied.

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Title:
Investigation of Filter Artifacts When Sampling Airborne Organic Particulates

Principal Investigator:
George J. Doyle

Institution:
University of
California-Riverside

Project Period:
2 years

Budget:
(2 years) $100,000

EPA Project Number:
R 808021

The objective of this research is to investigate the applicability of centrifugal preconcentration of air pollution problems, in particular the problem of reducing aerosol sampling artifacts. The plan is to demonstrate feasibility by building a small scale model of the preconcentrator operating at a sampling rate of 0.025 m^3/min and systematically studying its performance. If results are promising, further studies of the factors affecting performance will be carried out culminating in the construction of a preconcentrator sampling at rates useful for field sampling.

Work to date has consisted in design studies and in locating sources of materials needed to construct the bench scale preconcentrator.

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Title:
Heavy Metal Effects on Gene Expression in Human Cells

Principal Investigator:
Michael Karin

Institution:
University of California-San Francisco

Project Period:
2 years

Budget:
(2 years) $146,268

EPA Project Number:
R 809189

Metallothioneins are low molecular weight proteins which have unusually high cysteine content (30%) and no aromatic amino acids. Metallothioneins bind heavy metals that belong to groups Ia and Ib (Cu, Ag, Zn, Cd, and Hg). Their normal physiological role is probably regulation of trace element metabolism (particularly Cu and Zn). Since they are induced in a multitude of organisms (from yeast to man) by exposure to heavy metals, it has been proposed that they also have a role in protection against heavy metal poisoning.

Metallothioneins are induced in HeLa cells by glucocorticoid hormones as well as heavy metals. Both responses involve primary induction, probably at the level of transcription. This property of the gene(s), together with other properties, make the metallothionein system an attractive system for the study of regulation of gene expression.

The metallothioneins are small proteins and their gene is also relatively small (about 1 Kb in length) simplifying nucleic acid sequence analysis, an important prerequisite for understanding the molecular mechanisms involved in regulation of gene-expression.

Studying the regulation of metallothioneins expression in HeLa cells offers the opportunity of working with the best characterized cell line which exhibits high levels of induction—up to 50-fold with cadmium and ten-fold with glucocorticoids. Besides being a very valuable system for studying regulation of gene expression, the metallothione system offers the unique opportunity to study regulation of gene expression by metal ions. Some of these metals (such as cadmium) are potent carcinogens and teratogens and understanding their molecular interactions with the genome during induction of metallothione in transcription might help in understanding the basis for their action as carcinogens.

The research will pursue the following objectives:

(A) *Metallothionein Gene Structure*

- (1) Determine the nucleotide sequence of the human metallothionein genes and their 5′ flanking regions.
- (2) Obtain a genomic linkage map of the different metallothionein genes in humans.
- (3) Determine the chromosomal location of the human metallothionein genes by screening DNA from mouse X human cell hybrids, each of which contains a different human chromosome.

(B) *Determine the Nucleotide Sequences Important for Regulation of Metallothionein Expression by Heavy Metals and Glucocorticoids*

- (1) Look for nucleotide sequence homologies in the 5′ flanking regions of the metallothionein genes from different species (yeast and human).
- (2) Transfer the human metallothionein genes into mouse LTK$^-$ cells and determine the level of their expression and regulation. In the event they are expressed and regulated, a series of deletion mutants in the 5′ flanking regions will be prepared and a determination made whether those deletions affect the expression

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and regulation of the foreign metallothionein genes in the mouse L cells. Another approach will be to construct a chimeric plasmid that has the 5′ flanking sequence from a metallothionein gene and the coding region of the Herpes Simplex TK gene. This hybrid gene will be used to transform LTK⁻ cells which will then be examined for regulation of TK activity by heavy metals and glucocorticoids. Those chimeric plasmids have already been constructed.

(3) An alternate approach to gene transfer will be to construct a new hybrid plasmid that contains a metallothionein gene and part of the genome (sufficient for transformation) of Bovine Papilloma virus (BPV) DNA to transform primary cultures of mouse cells. BPV transforms mouse cells and exists as an autonomously replicating unit of low copy number. The regulation of the foreign metallothionein genes in such transformants will be studied.

(4) Analyze control regions of the metallothionein gene by *in vitro* transcription. The deletion mutants used for gene transfer experiments will be used as templates for *in vitro* transcription system that will respond to heavy metal ions and will be useful in analyzing control sequences different than the TATA box.

(C) *Analyze Fibroblasts Cultures from Patients with Genetic Defects in Trace Element Metabolism*

There are four genetic defects involving copper and zinc metabolism: Wilson's disease, Menke's Kinky hair syndrome, acrodermatytis enteropathica and familial hyperzycemia. Defects in metallothionein expression have been proposed as the cause for all of these syndromes. The availability of cDNA probes for the human structure and/or expression accounts for these inborn disorders of copper and zinc metabolism.

Some experiments directed toward this objective have been undertaken already. Results to date indicate that the induction of MT mRNA by different metals and glucocorticoids to be readily detected in primary human fibroblast cultures by Northern hybridization, as a first step toward molecular understanding of these defects.

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Title:
SCE Assay for Waterborne Genetic Hazards

Principal Investigator:
Michael B. Maddock

Institution:
College of Charleston

Project Period:
3 years

Budget:
(2 years) $168,080

EPA Project Number:
R 808251

Many chemicals that eventually enter aquatic ecosystems have potential genotoxic activity which may adversely affect populations of aquatic organisms. Aquatic species that serve as food sources could in turn serve as vectors for human exposure and possible increases in the incidence of genetic disease, birth defects and cancer.

Research is in progress to further develop, evaluate and apply to field situations short term assays to measure chemically induced genetic damage in an estuarine teleost, the oyster toadfish (*Opsanus tau*). The research design is based on a previous demonstration of sister chromatid exchange (SCE) induction in dividing toadfish leukocytes that were exposed *in vitro* to a chemical mutagen (ethylmethanesulfonate).

There are five objectives of this program:

(1) To evaluate the induction of sister chromatid exchanges and of chromosomal aberrations (CA) in cultured toadfish leukocytes as indicators of genetic damage following *in vitro* exposure to known mutagenic carcinogens.

(2) To develop and evaluate procedures to measure SCE and CA induction following *in vivo* exposure (ip injection) to known mutagenic carcinogens.

(3) To evaluate the sensitivity of *in vivo* induction of SCE and CA for detecting the genotoxic effects of waterborne mutagenic carcinogens.

(4) To examine the possibility that residual genetic damage in natural populations of toadfish can be detected as increased SCE rates in dividing leukocytes from freshly caught, untreated fish.

(5) To examine the possibility that *in vivo* assays for SCE and CA induction can be used for *in situ* assessment of genotoxic hazard from point sources of estuarine pollution.

Studies will also be continued to examine xenobiotic metabolism, DNA repair and *in vitro* cell transformation in fish tissues with the aim of developing further *in vivo* assays for waterborne genetic hazards.

Title:

Malignant Transformation of Human Diploid Cells by Chemical Carcinogens and Mutagens from Residues of Drinking Water

Principal Investigator:
Henryka J. Kurzepa

Institution:
University of Cincinnati

Project Period:
3 years

Budget:
(2 years) $245,124

EPA Project Number:
R 808703

The main goal of this research is to define a reliable human model *in vitro* system applicable to an assessment of environmental carcinogens. The approach is based upon our initial results using low-passage, normal diploid karyotype fibroblasts derived from newborn foreskin (AG1518). Features of this study will include alternate methods of exposure to the chemical agent, and examination of multiple criteria for recognition of the transformed phenotypel. Initial experiments will use N-methyl-N'-nitro-N nitrosoguanidine applied to the cells in doses which range from 1 to 15 g/ml.

Cells will be exposed according to two protocols. Monolayers will be treated for 24 hours, washed and cultivated through several doublings. These cultures will be examined for morphological and growth changes, for colony formation in semi-soft agar and for malignancy in nude mice. Mice will be innoculated by subcutaneous or intramuscular injection, or by surgical implants of pelleted cells imbedded in agar discs. Representative colonies in soft agar will also be propagated if possible for transplantation to nude mice. Alternatively, cells will be seeded in semi-soft agar (10^4 cells/well) and exposed 24 hours later to the carcinogen. These will be cultivated and monitored for colony formation for up to three weeks. Representative colonies will be isolated and tested for malignancy in nude mice, directly and after *in vitro* propagation. Tumors will be characterized histologically and for human cell markers (karyotype and biochemical markers). Subsequent experiments may include tests of multiple exposure to single agents. Successful protocols will be tested using other known carcinogens and environmental mutagens.

Title:
Biotransformation of PAHs by Aquatic Plants

Principal Investigator:
David Warshawsky

Institution:
University of Cincinnati

Project Period:
2 years

Budget:
(2 years) $299,992

EPA Project Number:
R 808459

The specific aims of this research using green algae, duckweed, and tobacco callus and model plant systems are to investigate the following:

(1) the metabolism of polycylic aromatic hydrocarbons (PAHs)—metabolism in light and dark condition will be compared and the possibility of metabolism by extracellular enzymes will be explored;
(2) the mutagenicity of the total organic extractable metabolic products—metabolite patterns and mutagenicity of total metabolites will be determined and compared to these parameters observed in mammals;
(3) the accumulation of PAHs—uptake of PAHs of varying structure (and biological activity in mammals) wil indicate if accumulation is influenced by structure; and
(4) the effects of PAHs on growth, chlorophyll content and energy stores (ATP).

Solubilized radiolabeled PAHs are diluted with unlabeled PAHs to achieve the desired concentrations. Aliquots of PAH solutions are added to plant cultures which are incubated either in the dark or in light-dark cycles with conditions optimal for growth of specific organisms. In relation to incubation in time and photoperiod, PAHs are extracted from plant tissues and media. Total PAHs and individual metabolites in plant tissue and media are quantified by high performance liquid chromatography and liquid scintillation spectrometry. The mutagenic potential of total metabolites (separated from the parent compound) is determined in the *Salmonella*/microsomal assay.

Three PAHs with differing ring structure and biological activity in mammals are used: benzo(a)pyrene, 5 rings, strong carcinogen; benz(a)anthracene, 4 rings, weak carcinogen and anthracene, 3 rings, noncarcinogen. Establishing a relationship between structure and uptake will aid in interpreting the results of Graf and Diehl (1966) and Payer *et al.* (1975), who reported that growth enhancement of algae was due to the selective accumulation of carcinogenic PAHs.

Metabolites of subtoxic concentrations of PAHs observed in growth media may be excreted after cellular metabolism or may be metabolized by extracellular enzymes secreted by plants. To identify the site of metabolism, oxidation products of PAHs are assayed from incubation of PAHs in media removed from cultures with and without prior PAH exposure. Medium for extracellular metabolisms studies is removed from plant cultures at various times related to growth.

This proposal contains new direction. Metabolism of PAHs by mammals and the biological effect of the metabolism, particularly BAP, has been studied, but little work has been done in plant systems. Although we can only hypothesize, at this point, as to the potential significance of PAH interactions with plants, it is important to extend our studies on PAHs in terms of their accumulation and metabolism, as well as their effects on growth of plants. It is critical to know if PAHs in soil, surface waters, sludge, and other wastes, are accumulated and metabolized to mutagenic compounds which must be considered in the assessment of risk.

Title:
Accelerated Settling of Suspensions by Addition of Buoyant Particles

Principal Investigator:
Ralph H. Weiland

Institution:
Clarkson College

Project Period:
2 years

Budget:
(2 years) $83,864

EPA Project Number:
R 808519

The objective of this research is to further develop the recent discovery that the settling rate of a suspension can be enhanced manyfold if a phase of opposite density to the settling materials (relative to the suspending medium) is added to the slurry. The proposed work will develop along two lines: (1) a detailed fundamental investigation of the *structure* of the convective motion set up by the presence of the extra phase, and (2) a full investigation on the use of a multitude of small *gas bubbles* as settling promoters. The latter will be done mainly through batch experiments, but with a brief amount of exploratory work on the continuous mode using a bench scale continuous settler. The treatment of liquid-liquid dispersions is also of interest and will be the subject of some preliminary experiments as part of Item (1) below. Specifically, we propose to do the following:

(1) Investigate the influence of physical properties such as density differences, polydisperse particles size, type of particles phase (regular and irregular shape solids as well as liquid droplets and gas bubbles) and vessel size on the *evolution* of uniform settling into cluster formation and intense streaming.

(2) Investigate the utility of large numbers of small gas bubbles as settling promoters by measuring disengagement and total settling times and determining the settling efficiency and optimum operating conditions for maximum enhancement. If *total* settling of a suspension is the objective, as opposed to partial thickening, which is distinctly different in three-phase settling (although not in conventional settling), then the use of *solid* particles as the added phase causes a number of problems. Not the least of these are (i) the necessity of recycling the added phase back to the start of the process, and (ii) the fact that what are optimal conditions for the fast removal of one of the species are far removed from the optimum for the other phase. The use of gas bubbles will form a major part of the work, quite apart from its connection to item (1) above.

(3) Perform some preliminary experiments on continuous settling using gas bubbles as the accelerating agent in a bench-scale or miniplant settler.

Title:
Basic Physical Properties of Pollution Control Residues

Principal Investigator:
Richard I. Dick

Institution:
Cornell University

Project Period:
2 years

Budget:
(2 years) $142,500

EPA Project Number:
R 808324

The physical properties of residues produced by air and water pollution control activities exert major influences on the gross volume of residues to be managed and on the cost of residue management. Procedures for design and operational control of processes whose performance depends on the physical characteristics of sludges developed, of necessity, from practice, and basic understanding of the performance of most of the processes still is lacking.

The objective of the proposed research is to develop means for characterizing the basic physical properties of sludges so as to improve the ability to effectively design and operate sludge management facilities. In recognition of the acquired value of conventional empirical measures of the physical properties of sludges, the significance of such parameters will be interpreted in terms of basic measures of the physical properties of sludges. The net result of successful completion of the research would be a reduction in the gross mass of sludge produced in the course of environmental quality control and a reduction in pollution control costs.

More specific major objectives of the proposed research are to:

(1) Develop means for characterizing the fundamental physical properties of sludges that influence the performance of sludge management processes. (Examples of fundamental physical properties include permeability, particle size, density and compressive strength.)
(2) Develop a body of information on the basic physical properties of sludges of varying types. This will be done using the techniques developed for the first objective.
(3) Determine the relationships between fundamental physical properties of sludge and conventional empirical measures of the physical properties of sludges. (Examples of conventional measures of physical properties of sludges include the sludge volume index, specific resistance, and capillary suction time.)
(4) Assess the limitations of conventional empirical measures of the physical properties of sludge in design and operation of facilities for sludge management.

Title:
Methane Fermentation of Coal Conversion Wastewater Constituents

Principal Investigators:
R. E. Speece
G. F. Parkin

Institution:
Drexel University

Project Period:
2 years

Budget:
(2 years) $168,000

EPA Project Number:
R 808010

Wastewaters generated from coal conversion processes are expected to contain phenolics, substituted phenolics, polycyclic aromatics, heterocyclics, and aromatic acids and aldehydes. These wastewaters will most likely be treated biologically, and there are several significant advantages to anaerobic biological treatment. Since the exact composition of coal conversion wastewaters (CCWW) is yet unknown and is expected to be variable, it is, therefore, desirable to assay the anaerobic biodegradability and toxicity of individual components expected to be present in CCWW. Such fundamental information will allow pollution control engineers to predict the potential degradability of a CCWW once its characteristics are determined.

The general objective of the research is to develop a catalogue of CCWW components which are amenable to methane fermentation. Each of the twenty-four compounds expected to be found in the highest concentration in CCWW will be individually assayed for toxicity and biodegradation to methane. The CCWW constituent concentration that results in 50% inhibition to unacclimated cultures will be determined. Detailed attention will be paid to proper acclimation of the methane fermentation cultures; the extent of acclimation will be determined. The potential for co-metabolism will be assayed. For those components amenable to methane fermentation, biological kinetic coefficients will be determined. Also, of those constituents biodegradable to methane, the intermediate metabolites will be analyzed by GC and GC-MS techniques. During the second year of the study, the synergistic effects of CCWW components will be assayed. Also, a synthetic mixture simulating a CCWW will be assayed for biodegradation to methane.

Serum bottles, submerged anaerobic filters, pH stats, and complete-mix reactors will be employed using acetate, propionate, and phenol enrichment cultures along with a seed digester fed a mixture of organic substrates. Disappearance of individual CCWW components will be monitored using appropriate GC techniques and methane production will also be quantified using a GC. Identification of intermediate metabolites will be attempted using GC and GC-MS techniques. Mass balances will be calculated. A distinction between the concentration of material in the liquid phase and that absorbed on the biomass will be made using appropriate extraction techniques.

Title:
Production, Culture, Food Habits and Impact of Hybrid Grass Carp

Principal Investigator:
Jerome V. Shireman

Institution:
University of Florida

Project Period:
2 years

Budget:
(2 years) $80,342

EPA Project Number:
R 807970

The objectives of this project are to:

(1) Evaluate culture techniques to raise hybrid grass carp to sufficient size for stocking in weed infested lakes.
(2) Determine food habits and consumption rates of fry, fingerling, and adult hybrids, and to determine how their food habits relate to grass carp and endemic species.

Culture techniques are currently being investigated in circular tanks and pond situations. Grass carp and hybrids are being cultured simultaneously in 1.23 meter diameter tanks, utilizing duckweed (*Lemna* sp.) chara (*Chara* sp.), filamentous algae, hydrilla (*Hydrila verticillata*) and trout pellets as food. Growth rates and consumption rates are monitored closely in these experiments.

Hybrids have also been stocked in one-acre ponds where growth rate and food habits are determined. Experimental ponds are fertilized with organic and inorganic fertilizer to encourage plant growth, both phytoplankton and macrophytes. Two ponds are being circulated with paddle wheels to improve oxygenation and water movement. Fry will be stocked in these ponds to determine if water circulation will improve survival and growth. Food selectivity of fry, hybrids and adult fish will be determined in pond situations where fish have a choice of phytoplankton, macrophytes, invertebrates and fish. Preliminary data indicate that hybrids, unlike grass carp, prefer shara.

Title:

Particle Deposition in the Wake of Charged Spheres

Principal Investigator:
Michael J. Matteson

Institution:
Georgia Institute of Technology

Project Period:
2 years

Budget:
(2 years) $92,444

EPA Project Number:
R 809470

Earlier work by Matteson and co-workers wherein aerosols were collected from gas streams moving around charged water droplets indicated the likelihood of high rates of deposition on the aft side of the droplet where flow conditions are not adequately represented by potential flow theory. Further tests were made with metal spheres and the aerosol deposition pattern observed with scanning electron microscopy. Results from these tests confirm that the developed wake has a definite influence on the collection pattern as well as the overall collection efficiency. Generally, minima were observed aft of the separation point at an angle at which the vortices flow tangentially to the sphere. Maxima appeared forward of the separation and advanced toward the forward stagnation point as the Reynolds Number increased and overall collection efficiency declined. At the higher levels of field intensity, the deposition profile appeared flat as one scanned around the sphere with a peak at the rear stagnation point. Very little information is available concerning deposition in the wake of moving droplets.

The collecting sphere's efficiency is highly dependent on its position relative to other spheres. Not only is the particle collection affected by the wake from leading spheres, but also by the electrical fields of neighboring spheres. This seems particularly important for scrubbers and spray towers where particles are collected by droplets moving at high Reynolds numbers where wakes and eddies are quite common.

The aim of the proposed work is to:

(1) Measure local particle deposition (deposition as a function of angle around the equator of the collecting sphere) for various size collecting spheres and particles, particle charge and field strength, and relative velocities.

(2) Measure particles deposition in an array of charged spheres with various relative distances and orientations between spheres and various relative velocities, charge and field strength, particle and sphere size.

(3) Correlate particle deposition with Reynolds, Sotkes', Coulomb attraction parameters, and orientation within a field of charged spheres.

The initial experiments are to be conducted with brass spheres in the 0.1-6.0 mm diameter range. Particles will consist of pre-washed, latex spheres in the 0.1-5.0 micrometer range. The particles will be dispersed with a DeVilbiss No. 40 nebulizer, dried, and then charged in a concentric wire-in-cylinder corona. Particles will be analyzed to assess the effects of residue aerosol. The flowing aerosol stream will impinge on a sphere mounted in the center of the flow channel. The sphere is removed, mounted on a rotatable scanning electron microscope stub, and photographed at various positions around the equator.

In the second phase of the experimental work, a series of collecting spheres will be mounted in a line along the axis of flow. In this arrangement, the deposition on each sphere can be compared with one another to ascertain the relative efficiency with order in line. In another set of tests, the spheres will be staggered in an array, much like bowling pins, and set first at one distance to one another and then another. A final series of tests will be made with a three-dimensional array with both a square and staggered configuration of collectors.

Title:
Soot Formation in Gaseous Diffusion Flames

Principal Investigator:
J. I. Jagoda

Institution:
Georgia Institute of Technology

Project Period:
2 years

Budget:
(2 years) $97,381

EPA Project Number:
R 808953

Soot released during hydrocarbon combustion in fires and many combustion devices is a major health hazard as well as a technical problem due to the increased heat transfer to the surroundings. In order to facilitate a better understanding of soot suppression techniques (e.g., by additives) the mechanisms of soot formation in a diffusion flame is being investigated. Smoke, i.e., aggregated soot particles, passes through four stages during its formation: pyrolysis of the original fuel, nucleation, growth by surface deposition and/or coagulation and agglomeration. The physical mechanism for each of these steps must be known if the chain leading to soot formation is to be successfully broken.

Local soot particle diameters and soot mass loadings are being measured using suction probe techniques and transmission electron microscopy. The sizes and number densities of soot aggregates and soot volume loadings are determined using simultaneous laser light scattering and absorption. Mapping out these soot particle and agglomerate characteristics as well as temperature and ion concentrations (Langmuir probe) throughout the flame will result in the determination of the position and, therefore, the environment in which the individual steps of soot formation occur. This will allow the investigators to suggest which of the mechanisms found in the literature for each of these steps is appropriate to the diffusion flame.

Recent advances in the field have suggested that flame temperatures largely determine the amounts of soot found in flames. The above measurements are, therefore, carried out in a diffusion flame whose temperature is varied by adding varying quantities of inert gas, such as nitrogen, to the fuel flow. This will permit the determination of the extent of the effect of temperature on the different soot formation steps outlined in the first paragraph.

The main fuel used in this investigation is propane. Tests will also be run using aromatics and gases consisting of conjugated molecules, such as butadiene, in order to determine the effect of the molecular structure of the fuel on the formation of soot.

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Title:

A Project to Develop Emission Spectroscopic Methods for Determining the Halogens and Other Nonmetals Via Direct Current Plasma Excitation

Principal Investigator:
J. Taylor

Institution:
Hampton Institute

Project Period:
3 years

Budget:
(2 years) $81,386

EPA Project Number:
R 808676

The purpose of this project is to develop atomic spectroscopic methods for the determinations of nonmetallic elements using direct current plasma as the excitation medium. The elements of interest in this work include the halogens, phosphorus, silicon, arsenic carbon, boron, sulfur and selenium.

Measurement methods will be developed to determine chlorine, fluorine, and bromine in the form of aluminum monohalides. Analytical data for these elements will be obtained with the d.c. plasma system as well as with an atomic absorption unit. These two sets of data will be compared with respect to detection limits, linear dynamic range, precision, and other parameters to evaluate the applicability of these approaches to analytical measurements.

Atomic emission methodologies, via d.c. plasma excitation, will be developed and characterized for carbon, boron, phosphorus, silicon, sulfur, arsenic and selenium. The characterization of the developed methodologies will be done on the basis of achievable sensitivities, detection limits, analytical range, and interference effects. The methodologies developed here will be compared with other traditional and nontraditional methods which have been employed for the determination of these elements.

Both chemical and spectral interference effects will be evaluated. The means of introducing samples into the plasma system will be of prime concern. Thus, several nebulizer systems will be evaluated including Bobington, Meinhard, and crossflow nebulizers.

The methodologies developed in this work will be applied in the determination of cited nonmetals in environmental samples including natural waters and wastewaters. Samples will be collected from several locations in the Hampton Roads Area. These locations will include natural fresh water reservoirs and creeks as well as wastewater treatment plant effluents and industrial discharges. In some cases, samples will be obtained from other laboratories which are engaged in the determinations of the elements in question. This will serve as cross-comparison of the methods developed in this work with those which are being used by others.

Further evaluation of this work will rely on the comparison of the data obtained with those which have been reported in the literature using other methods. In some instances, e.g., the determination of phosphorus and silica, parallel measurements will be conducted for direct comparison of the methods developed here with other conventional ones such as uv-vis spectrophotometry.

This project will be conducted in two phases. The first phase will constitute methods development and evaluation. The second phase will entail the applications of the methodologies developed to the analysis of environmental samples.

Title:
Venturi Scrubber Performance and Optimization

Principal Investigator:
David Leith

Institution:
Harvard School of Public Health

Project Period:
3 years

Budget:
(2 years) $223,995

EPA Project Number:
R 809003

Venturi scrubbers are widely used for control of fine particles from industrial processes. A major cost in control by scrubbing is due to energy consumption from high gas-phase pressure drop.

Recent theoretical work has shown that venturi performance can be optimized through atomization of drops with proper size. This can be accomplished by selection of proper gas velocity and liquid injection rates through the venturi throat. Theoretically, venturi optimization allows a substantial reduction in pressure drop, and thus reduces energy consumption and operating cost in comparison with a conventional venturi operating at the same efficiency. Alternatively, a substantial improvement in efficiency is theoretically possible with an optimized venturi, in comparison with a conventional venturi operating at the same pressure drop.

Experimental data from the literature, developed for another purpose, support these theoretical conclusions. However, careful experimental work is necessary to confirm this theory directly, to improve it, and to establish guidelines for its practical use.

The objectives of the present work are to:

(1) Develop data that allow further refinement of venturi performance and optimization theory.
(2) Demonstrate the extent to which the theoretical benefit from venturi optimization is achieved.
(3) Develop recommendations for venturi design and operation so that these benefits can be utilized in a practical way.

Implementation of results from this project may save as much as one-third of operating cost for venturi scrubbers already operating. Even greater savings may be possible with new venturis designed according to guidelines to be developed at the Harvard School of Public Health. These savings are particularly important in light of current high costs for energy.

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Title:
Limitations and Energetics of Hydrophobic (Nonpolar) Sorption of Organic Compounds by Soils

Principal Investigator:
John J. Hassett

Institution:
University of Illinois

Project Period:
3 years

Budget:
(2 years) $144,438

EPA Project Number:
R 808417

The objectives of the proposed research are:

(1) To examine the mechanism(s) involved in the hydrophobic sorption of nonpolar organic compounds by soils and sediments. Sorption constants are being measured in different solvent systems and at different temperatures in an attempt to measure the enthalpies and entropies of sorption. Hydrophobic sorption appears to be an entropy driven process and these measurements should provide insight into the driving mechanisms. Both batch equilibrium sorption experiments and kinetic experiments are being conducted.

(2) To determine the limitations of the log Koc-log Kow and log Koc-log S relationships that have been developed by several investigators to allow prediction of a compound's sorption constant (Koc) which has been normalized for the organic carbon content of the oils or sediment from the organic compounds octanol-water partition coefficient (Kow) and/or its water solubility (S). Soils and sediments with a wide range of pertinent properties have been collected from a wide geograhic area. These soils will be used in sorption experiments with a wide range of organic compounds to define the limits of the log Koc-log Kow and log Koc-log S relationships. High pressure liquid chromotography is being used to determine the concentration of the compounds in the sorption experiments.

Title:
Effects of Toxic Substances on Memory

Principal Investigator:
George A. Heise

Institution:
Indiana University

Project Period:
3 years

Budget:
(2 years) $194,916

EPA Project Number:
R 809152

The research is a comparison of the effects of exposure to toxic substances and reference drugs on memory, acquisition, and general performance of young, mature, and old rats. In addition to providing useful data on particular toxic substances, the research has the more general objective of establishing the feasibility and importance for behavioral toxicology of determining effects of toxic substances on memory and acquirisition, and of demonstrating procedures by which this can be accomplished.

There are four related goals:

(1) *Introduction of new methods for behavioral toxicology.* Rats will be trained and tested in four discrete-trail operant procedures developed in our laboratory for measuring memory acquisition, and general behavior. These methods provide stable baselines of behavior that have been shown to be sensitive to drug treatment and should also be sensitive to toxic substances. The various measures of memory will be compared with each other and with other various measures of behavior from the same and other situations for sensitivity to the toxicants and reference drugs, and for analyzability and general feasibility.

(2) *Determination of effects of particular toxicants on memory, acquisition and general behavior.* Initially, this research will determine the effects on behavior in the four test situations of three insecticides that affect cholinergic system functioning: carbaryl (1-naptyl N-methyl carbamate), Baygon (Propoxur; o-isopropophenyl N-methyl carbamate) and Decamethrin, a synthetic pyrethroid. Carbaryl and Baygon inhibit cholinesterase (Goldberg, 1983; Desi, 1974) and decamethrin has some cholinergic-like effects on behavior (Ray and Cremer, 1979). The toxicants will be compared for behavioral effects with three reference drugs: scopolamine, and anticholinergic; phsostigmine, an anticholinesterase; and d-amphetamine, which is catecholaminergic in behavioral effect at the doses to be used in this research. Later, other toxic substances will be studied, using optimal methods and procedures developed in the research with toxicants named above.

(3) *Evaluation of various treatment regimens.* Sensitivity to toxicants is expected to depend on age of the animals and the conditions of treatment. In order to evaluate these effects, acute toxicants will be administered to rats of three different ages; beginning at <30 days, 70, and 470 days of age. Animals will be trained and tested with toxicants and reference drugs at the latter two stages. In addition, some animals will receive subacute administrations of a toxicant during the period between the testing stages.

(4) *Enhanced understanding of behavioral and biochemical processes in animal memory.* The four behavioral situations permit separation of treatment effects on memory performance from effects on other behaviors. The role of underlying neurotransmitter processes can be inferred from known pharmacological action of the toxicant and reference drugs used.

Title:
Fundamental Analytical and Spectroscopic Studies Exploiting Laser Ionization

Principal Investigator:
James P. Reilly

Institution:
Indiana University

Project Period:
2 years

Budget:
(2 years) $163,490

EPA Project Number:
R 808790

The conjunction of laser-induced molecular ionization with mass spectrometry is particularly fortuitous since it enables us to learn more about the former phenomenon by exploiting the latter technique; while at the same time we may be able to improve the latter technique by exploiting some of the unique properties of the former phenomenon. One of its most significant features is the potential impact that multiphoton ionization mass spectrometry (MPI/MS) may have in analytical chemistry. In order to better measure this, the Principal Investigator is continuing his investigations of the ionization probability of a variety of organic compounds.

The substances chosen are industrial products manufactured in massive quantities which now appear at barely detectable levels in our environment. Experiments of a fundamental nature will also be performed with the proposed apparatus to probe more deeply into the ionization phenomenon. The low-pressure, collision-free operating conditions, the excellent time resolution provided by available pulsed discharge lasers and the sensitivity of the technique facilitate novel studies of the interaction of laser radiation with isolated gas phase molecules. Molecular spectroscopic, excited state relaxation and ionization potential data are all extractable in the proposed experiments. In some cases, this information can even be obtained on transient species.

The Principal Investigator proposes to set up a general purpose apparatus which is a combination of two already successfully operating spectrometers.

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Title:
Evaluation of Factors Affecting Selection of Media for Anaerobic Packed-Bed Reactors

Principal Investigator:
James C. Young

Institution:
Iowa State University

Project Period:
2 years

Budget:
(2 years) $137,030

EPA Project Number:
R 809524

The advantages of using anaerobic PBRs for treating low-strength soluble organic wastes effectively and efficiently at nominal temperatures have been demonstrated. However, previous in-depth investigations have been limited to small laboratory-scale units and to the use of small plastic, stone or ceramic modules as a packing medium. Larger units have been constructed for treating some industrial wastewaters, but the designs often were based on limited data, and their long-term performance in some cases has been less than expected.

One important design factor about which little is known is the effect of media shape and size. A computer model suggests that the critical factors affecting PBR performance—biological solids entrapment, biological solids transport, surface area of attached and flocculated solids, and short-circuiting—are related to media design and size. Both the need for and frequency of biological solids wasting no doubt is related to media design and size, as is the potential for plugging.

Media design and size likely will be dictated by intended application. For example, units containing small media generally are expected to give better waste removal performance than units containing large media when operating at the same loading rate. Consequently, scale-up using parameters developed from test units containing small media to the designing of units using large media could introduce considerable error.

Because of these needs, the subject research study involves operation of a number of anaerobic PBRs under controlled conditions for the purpose of achieving the following objectives:

(1) Determining the effect on PBR performance of using different sizes and designs of media with the goal of optimizing gas production and waste removal through proper selection of media.
(2) Determining the effect of media design and size on biological solids accumulation and the need for solids wasting with the goal of achieving long-term satisfactory operation with a minimum of sludge wasting and reactor cleaning.
(3) Determining the relationship between organic loading, performance, and media shape and size with the goal of identifying design parameters for process scale-up.
(4) Refining mathematical models to obtain a better understanding of the fundamental operation of the system and to help identify parameters of importance to design, particularly those related to media design.

All tests will be conducted using pilot-scale reactors 20-inches (0.5m) in diameter and 6 feet (2m) tall. Reconstituted industrial wastes will be fed to these units in various concentrations and over a wide range of organic loadings to determine the media-related loading limits and performance characteristics.

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Title:

Metabolic Factors as Determinants of Lung Toxicity

Principal Investigator:
David J. P. Bassett

Institution:
Johns Hopkins University

Project Period:
3 years

Budget:
(2 years) $163,127

EPA Project Number:
R 809091

This study will provide the basis of understanding those factors that affect the retention, clearance and health consequences of inhaled substances found in the environment.

The combustion of fossil fuels, such as coal and oil, results in the generation of air pollutants that include polycyclic aromatic hydrocarbons (PAH) such as the carcinogen benzo(a)pyrene, and the oxides of nitrogen and sulphur. Assessment of risk to human health caused by PAH emissions requires an understanding of the metabolic and environmental factors that influence their deposition, retention and clearance in lung tissue. Since the alveolar region represents a major deposition site of PAHs bound to submicron particulates, an isolated perfused rat lung preparation will be used to characterize the role of lung parenchymal tissue in the activation and detoxification of inhaled PAHs. Such metabolism will influence the amount of deposited PAHs available for translocation to airway sites of PAH-induced lung cancers.

A model system for evaluating lung pathways of PAH metabolism will be established, based on benzo(a)pyrene (BaP) administered to perfused rat lungs either in the perfusate or by tracheal instillation. BaP metabolism will be measured by radioisotope incorporation into primary and secondary metabolites, isolated from the perfusate and lung tissue fractions at different times of perfusion. Since the various activation and detoxification pathways require the coenzymes NADPH and reduced glutathione (GSH), the metabolic control of these pathways will be investigated by the use of inhibitors of GSH metabolism and by perfusion with electron acceptors such as methyl viologen (paraquat) and phenazine methosulphate, that affect the supply and redox-state of NADPH. This system will then be used to evaluate changes in the pattern of PAH metabolites and enzymes in lungs taken from animals pretreated with carcinogenic and non-carcinogenic PAHs. Since the tissue supplies of NADPH and GSH are also required for cellular *de novo* fatty acid synthesis and anti-oxidant defense mechanisms, relationship between PAH exposures and these lung intermediary metabolic processes are, therefore, to be established. Chronic exposures to oxidant pollutants have been shown to alter the lung enzymes of NADPH and GSH metabolism. The pathways of PAH metabolism will, therefore, also be evaluated in lungs taken from animals following exposures to ozone.

Title:

Neurotoxicity of Low Level Prenatal Exposure to Hypoxic Agents

Principal Investigator:
Laurence D. Fechter

Institution:
Johns Hopkins
University

Project Period:
3 years

Budget:
(2 years) $142,002

EPA Project Number:
R 809061

Carbon monoxide (CO) is a major environmental health hazard for which occupational and ambient air exposure standards have been promulgated. The data upon which these standards are based comes largely from fully developed adult organisms living at sea level. Far less data is available on potential high risk subgroups such as the aged, chronically ill or immature organism or individuals residing at high altitude (c.f. National Commission on Air Quality report, "Health Effects of Carbon Monoxide at High Altitude," Denver, 1980). Based on a variety of physiological considerations including normal fetal oxygen consumption and availability of adaptive responses of hypoxia, there is reason to believe that the fetus is particularly sensitive to CO and presumably other chemical asphyxiants such as dichloromethane (DCM).

The Principal Investigator has recently described a variety of functional deficits produced in rats exposed prenatally to a low concentration of CO and has begun to determine the neurological basis for these toxicity signs. Additional experiments with CO will be conducted to determine a threshold level at which toxicity occurs and whether such toxicity can be attributed to exposure during a sensitive or "critical" gestational period. Parallel experiments with DCM will be used to help predict the generality of the findings with CO to other chemical asphyxiants. These studies will also provide basic information concerning the toxicity of DCM. The specific measures of toxicity to be used include tests of behavioral development, learning and retention and biochemical measures of nucleic acid and protein levels in brain regions.

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Title:

Detection of Somatic Cell Mutations in Human Erythrocytes by Labelling with Antibodies Against Glycophorin A

Principal Investigator:
Ronald H. Jensen

Institution:
Lawrence Livermore National Laboratory

Project Period:
3 years

Budget:
(2 years) $295,436

EPA Project Number:
R 808642

The principal objective of this project is to develop and test an assay for detecting somatic cell mutations in human erythrocytes. The assay is based on labelling cells taken from peripheral blood of humans with monospecific monoclonal antibodies against the surface glycoprotein, glycophorin A. Monoclonal antibodies to each of the two allelic forms of glycophorin, glycophorin A^M and glycophorin A^N are being isolated and conjugated with different fluorescent labels. Erythrocytes from individuals who are heterozygous at the glycophorin locus contain both allelic forms on the cell surface. Thus, a large majority of these red cells will be doubly labelled. However, stem cells that were mutated at the glycophorin gene should throw off daughter cells that would contain only one of the two allelic forms on their surface and thus would be only singly labelled. Dual beam flow cytometry of the labelled cells should allow enumeration of these rare, putatively mutant cells.

The frequency of such cells should be a measure of accumulated genetic injury to the erythroid stem cells of individual humans and therefore permit the early effect of mutagenic exposure to be detected. Genetic biochemical studies also will be performed to validate that the assay detects somatically mutant cells and that the frequency of such cells is related to past exposure of the individual to mutagenic chemicals.

Title:

Rapid Screening Method for Genotoxic Chemicals

Principal Investigator:
Larry H. Thompson

Institution:
Lawrence Livermore
National Laboratory

Project Period:
3 years

Budget:
(2 years) $258,314

EPA Project Number:
R 808641

Using mammalian cells, a unique short-term assay that can efficiently screen for chemical mutagens will be developed. The test employs mutant strains of Chinese hamster ovary (CHO) cells that are substantially more sensitive than the parental wild type (WT) to killing and mutation induction by a broad spectrum of mutagens. Different mutant strains show specificity in their sensitivity to mutagen classes, as the result of different *defects* in the *repair* of damaged DNA. Compared to human repair-deficient mutants such as xeroderma pigmentosum, the CHO strains have much more favorable growth properties, derive from a common parent, and can likely provide a broader spectrum of hypersensitivity to mutagens. This assay will exploit cytotoxicity measurements, which are easily and rapidly performed, and will detect genetic damage as differential killing of WT vs. mutant cells. This difference provides a simple indication of unrepaired lethal damage to the DNA of one or more sensitive strains. Assays will be conducted in 24-well plastic trays in which the cells are exposed to mutagen for 4 hours, incubated for 3 days for cell growth, and then fixed and stained. The intensity of staining is a measure of the amount of cell growth and can be converted into a semiquantitative inverse measure for toxicity.

The genetic significance of the differential cytotoxicity will be confirmed on selected compounds by demonstrating enhanced forward mutation induction in the repair-deficient cells, using two drug resistance markers (asaadenine resistance and thioguanine resistance). At the same time, the utility of repair-deficient cells for standard mutagenicity tests will be assessed. The most appropriate mutant strains will be selected and characterized in terms of sensitivity to different kinds of direct-acting mutagens (simple alkylating agents, large-adduct alkylating agents, and DNA cross-linkers), as well as promutagens requiring activation. Major emphasis will be placed on achieving efficient conditions for detecting a wide variety of promutagens (polycyclic aromatic hydrocarbons, aromatic amines, etc.). This will require defining the appropriate metabolic activation systems in the form of liver microsomal fractions or feeder layers of metabolically competent cells, such as hepatocytes. Standardized assays for cytotoxicity and mutagenicity will thus be established and applied to sets of known mutagens, as well as novel compounds of interest.

Title:

Reduction of Airborne Radon in Houses by Removal from Household Water Supply

Principal Investigators:
Jerry D. Lowry,
Willem F. Brutsaert

Institution:
University of
Maine-Orono

Project Period:
1 year

Budget:
(2 years) $64,293

EPA Project Number:
R 808724

The occurrence of radon in domestic water supply systems has been well documented. High concentrations have been reported in Maine, New Hampshire, and other New England states, North Carolina and Europe. Recently, the highest known radon concentration in a domestic water supply was measured at approximately 2.0 million picoCuries/L in Maine, where it is not uncommon to find Rn-222 levels in excess of 100,000 pCi/L in granite aquifers.

Radon gas is known to be carcinogenic by two pathways. In the first, radon is ingested with water directly, and in the second, radon is inhaled after partitioning into indoor air from radon-enriched water. The potential seriousness of the problem is growing due to the increasing number of drilled wells and the current trend toward well-insulated homes. Because of the health hazard related to airborne radon, the U.S. Surgeon-General, HUD, and the U.S. Environmental Protection Agency have recommended or are considering maximum working levels for radon concentrations in homes located in areas of high radon. Based on the strong correlation between radon in water supplies and in indoor air, it follows that removal of radon from water supplies must occur to avoid ingestion and inhalation in the household.

The intended project scope is comprised of two basic phases:

(1) Phase I: Laboratory Demonstration of Granular Activated Carbon (GAC) and Diffused Aeration Radon Removal Systems, and
(2) Phase II: Field Demonstration of Optimized Removal Units.

Phase I of the project also includes a theoretical study of the adsorption/decay phenomenon of radon removal by GAC adsorption, and Phase II encompasses indoor air quality monitoring of radon with and without prior removal from the water supply.

The objectives of Phase I will be accomplished by conducting the following research:

(1) Design and construct a radon generator to supply 350 gpd at a 100,000 pCi/L radon concentration.
(2) Extended period steady-state testing of four commercial GAC removal units (1.0, 1.5, 2.0, and 2.5 ft^3 capacity).
(3) Theoretical GAC adsorption/decay model development and experimental verification.
(4) Extended period testing of three diffused aeration units (40, 80, and 120 gallon capacity).
(5) Optimization of engineering parameters involved with the diffused aeration units.

Except for the theoretical GAC experiments, all removal research will be conducted using full-scale units, making a direct transition from laboratory research to field demonstration possible. It is anticipated that in the extended period of field research to follow, three GAC units and two diffused aeration units will be monitored in homes having water supply radon concentrations from approximately 30,000 pCi/L to greater than 100,000 pCi/L.

Title:

Toxicological and Chemical Interactions of Metal Oxides and Sulfur Dioxide

Principal Investigators:
Mary O. Amdur,
John F. Elliott

Institution:
Massachusetts
Institute of
Technology

Project Period:
4 years

Budget:
(2 years) $250,627

EPA Project Number:
R 809104

This is an interdisciplinary program, the main aim of which is to correlate the toxicological response of experimental animals exposed to metal oxide aerosols and sulfur dioxide with the nature of the particulate sulfur species formed. The experimental protocol permits the mixing of the metal oxides at chamber temperature or at furnace temperature. Water vapor may be added either to the exposure chamber or to the furnace so that the effect of humidity can be studied under different mixing temperatures.

The methods of toxicological assessment include measurement of mechanics of respiration in guinea pigs made before, during, and after exposure. Also measured are the uptake, clearance, and biological distribution of the metal oxide aerosols as affected by particle shape and by combination with sulfur dioxide. Other related work (supported by NIEHS) includes pulmonary function tests to evaluate post-exposure effects (lung volumes, static compliance, CO single breath diffusing capacity) as well as pathological studies. Thus, the biological response to the same experimental atmospheres can be evaluted in a variety of ways.

Current animal work uses combustion-generated zinc oxide aerosols, both as chain aggregates and as acicular particles, both in the submicron particle size range. Methods for determination of the nature of particulate sulfur species include Electron Spectroscopy for Chemical Analysis (ESCA), and specific wet chemical methods (West-Gaeke for sulfite and turbidimetric for sulfate). Development work on aerosol generation is currently a fine silica aerosol which will be surface layered with zinc.

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Title:
Peroxidase for the Removal of Toxic Pollutants from Industrial Waste Waters

Principal Investigators:
Alexander M.
Klibanov

Institution:
Massachusetts
Institute of
Technology

Project Period:
1 year

Budget:
(1 year) $47,313

EPA Project Number:
R 808619

The project deals with the application of the enzyme peroxidase from horseradish for the removal of toxic phenols and aromatic amines from industrial aqueous effluents. This approach is based on the phenomenon that peroxadase catalyzes oxidation of a number of different phenols and aromatic amines with hydrogen peroxide. As a result of this oxidation, water-insoluble polyaromatic products are formed. In contrast to their water-soluble precursors, these polymers can be relatively easily removed from water by such methods as sedimentation, filtration, flocculation, or coagulation. Therefore, in essence, the enzyme precipitates pollutants from water.

The enzymatic removal of phenols and aromatic amines from water will be studied quantitatively as a function of the concentration of peroxidase, H_2O_2, and pollutants and also of pH, temperature and treatment time. The mechanism by which easily removable phenols and aromatic amines aid in the removal of other pollutants by peroxidase and hydrogen peroxide will be investigated. The optimal operational conditions for the enzymatic treatment will be established. Experiments with "real" industrial aqueous effluents obtained from a chemical plant will be carried out. The economic feasibility of the peroxidase removal of hazardous pollutants from industrial waste water will be assessed.

Title:

The Formation of Carbonaceous Particulates During the Pyrolysis and Oxidation of Coals and Residual Oils

Principal Investigator:
Adel F. Sarofim

Institution:
Massachusetts Institute of Technology

Project Period:
2 years

Budget:
(2 years) $206,342

EPA Project Number:
R 808774

The purpose of this project is to systematically examine the influence of fuel combustion conditions and fuel composition on the formation of soot during the combustion of oils and coals and the formation of coked particles during the combustion of heavy fuel oil. Experiments are to be conducted on a finely dispersed stream of coal and oil particles entrained in a laminar flow of gas, the composition and temperature of which can be systematically varied. The carbonaceous particulate will be measured by the time resolved collection of the products of combustion and size classification of the particulate by aerodynamic means. Measurement of the soot concentration will also be made *in-situ* by monitoring absorption and emission by the particle cloud.

The variables to be studied are, for the coals, the particle size, coal rank, oxygen concentration of the ambient gas with particular attention to the role of volatile composition and mode of combustion on soot formation. Similarly for the oils the effect of droplet composition, droplet size and combustion environment on the soot formation will be studied with special attention on the effect of the extinction of the volatile flame by either a decrease in oxygen concentration or increase in CO_2 concentration. Multicomponent fuels will be used to examine the conditions under which coked cenospheres are produced as a consequence of the liquid phase cracking of heavy components and to the conditions under which droplet fragmentation occurs due to the internal boiling of high components. A model of the governing processes will be developed to facilitate the generalization of the results.

Title:
Ulva lactuca L. *as a Bioindicator of Coastal Water Quality*

Principal Investigator:
R. Wilce

Institution:
University of Massachusetts

Project Period:
15 months

Budget:
(1¼ year) $58,815

EPA Project Number:
R 808770

The principal objectives of this proposal are to test our preliminary results and refine methodologies concerning the use of green seaweed *Ulva lactuca* (sea lettuce) as a monitor of pollutants in coastal waters. Extracts from *Ulva* tissue occurring naturally and from deployed plants of uniform genotype will be used to screen for mutagens, organohalides and heavy metals. Procedures developed during the tenure of a Massachusetts Water Resources Grant will be combined with mutagenicity assay techniques to identify high priority populations of *U. lactuca* for subsequent intensive chemical investigation.

Our results using the *Escherichia coli* Pol A assay demonstrate that some *Ulva* populations are mutagen free; others have accumulated mutagenic compounds. Additional assays (Ames Test, *Saccharomyces cerevisiae* D4 and D7 strains) are proposed as confirmatory tests of the *E. coli* assays used to date. *Ulva* populations which give the strongest indication of mutagen accumulation will receive intensive chemical investigation.

Genotypically identical *Ulva* germlings will be deployed at selected statons, retrieved when mature *in situ*, and subjected to mutagenicity screening and tissue analysis. Naturally occurring *Ulva* plants from the same localities, and water samples, will be collected and analyzed for the compounds of interest. A wide range of stations will be designated, ranging from heavily polluted to chemically "clean," and will include a wide spectrum of physical regimes. Sufficiently numerous tissue collections will be made to permit conclusions concerning the variation of *Ulva* cell constituents over time. Efforts will be made to improve experimental protocols for both tissue analyses and germling deployment procedures.

Title:

The Testing of Fetuses and Newborns for Cardiovascular Problems after Exposure to Pesticides and Other Toxic Substances

Principal Investigator:
Casimer T. Grabowski

Institution:
University of Miami

Project Period:
3 years

Budget:
(2 years) $118,793

EPA Project Number:
R 808938

An urgent need in the field of teratogenic screening is the development of more sensitive methods for evaluating developmental toxicity. An ideal test or series of tests would provide (1) some objective evaluation of the general state of health and viability of fetus and newborns, (2) criteria which can be standardized so that the results obtained in different laboratories could be compared, and (3) reasonable economy in terms of operator time and equipment. A technique recently developed at the University of Miami, fetal and neonatal electrocardiography, meets these criteria and has potential for general use.

This technique was applied to study the effects of prenatal exposure to the fire ant pesticide, Mirex. This insecticide does not induce many visible abnormalities, but does precipitate a high incidence of perinatal mortality for totally unknown reasons. It was found that treated fetuses had heart blocks and other cardiovascular problems. Thus, it has been demonstrated that one of the major causes of the perinatal mortality induced by Mirex are functional problems with the cardiovascular system, something which standard teratological testing could not have determined.

The principal objective of this project is to develop fetal and neonatal electrocardiography of small mammals as standardized tests for evaluating developmental toxicity. This test can be an excellent diagnostic indicator of fetal health, just as useful as its human counterpart. Since it is based on physiological criteria, it has the potential of being more sensitive than those based solely on morphological criteria and should be a useful adjunct to traditional screening methods.

Another objective of this project is to evaluate the developmental toxicity of several pesticides, herbicides, and other toxic substances by studying the cardiovascular physiology of embryos, fetuses, and newborns prenatally exposed to them. The ECG test will be applied to rats and mice, especially at minimally toxic doses, to determine its usefulness and sensitivity to detect toxicity.

A subordinate goal is to apply this and other tests to study the physiological mechanism of action of Mirex and other pesticides on rat fetuses.

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Title:
Chemical Behavior of Chromium: Water-Sediment Systems

Principal Investigator:
David T. Long

Institution:
Michigan State
University

Project Period:
2 years

Budget:
(2 years) $62,721

EPA Project Number:
R 808306

Objective: The mobility of any metal in exogenic environments is controlled by adsorption and precipitation-dissolution reactions. These properties, in turn, are controlled by the environmental parameters Eh, pH, ionic strength, temperature, and composition of the solution and by the presence of charged substrates such as clays, iron-manganese oxides and organic material. The importance of adsorption on the mobility of metals is known, but the relative importance of the various adsorbing substrates in natural systems is not well known. This is particularly true for chromium. It appears, for example, that the relative importance of the listed substrates in controlling the migration of chromium is different among study areas and can have changes in their relative importances within a study area. The objective of this research is to gain a better understanding of the controls on the adsorption of chromium on various substrates. The working hypothesis of this research is that the differences in the relative importance of the various adsorbing substrates among and within study areas is a function of the complex interplay of the environmental parameters listed, the pH_{zpc} of the specific substrate, and the oxidation state of chromium. This objective will be accomplished by studying the fate of chromium in surface environments due to its addition from a highly industrial/populated area. The field study area is the Grand River, Lansing, Michigan.

Approach: The approach of this study is to determine the chemical partitioning of chromium in the stream sediments and interpret the results in light of the working hypothesis. Chromium concentrations in six fractions of the stream sediment (clays, iron oxides, manganese oxides, carbonates, organic matter, and detrital) will be determined by the method of sequential selective chemical attacks. The concentrations of Fe, Mn, Pb, Cu, and Zn will also be measured in these fractions and compared to the Cr concentrations to aid in the chemical interpretations. Chromium partitioning in the sediment as a function of grain size ($<$ and $>$ 0.15mm) will also be determined. A limitation of past work on determining the partitioning of chromium in sediments and soils is that few considered the effect of the amount of the adsorbing phase present when interpreting the importance of a particular phase in sequestering a metal. For some studies, not considering the amount of phase present is valid. On the other hand, studies that are trying to determine the relative importance of the phases and interpret the results chemically, such as in this study, this type of an approach is not valid. This study will consider the amount of adsorbing phase present in the interpretation of the results by using the concept of phase concentration factor which attempts to normalize the partitioning results to equal amounts of phase present. The partitioning results will also be studied without normalizing in order to determine what effects this may have on the interpretation of the data.

Title:

Development and Application of an In Vitro *Tumor Promoter*

Principal Investigator:
James E. Trosko

Institution:
Michigan State University

Project Period:
3 years

Budget:
(1 year) $261,623

EPA Project Number:
R 808587

Carcinogenesis appears to be comprised of two distinct phases, namely *initiation* and *promotion*. The initition phase describes the permanent alteration of a single cell to a "premalignant" or initiated status. Agents which damage DNA or mutate cells appear to be initiators. Promotion involves the clonal expansion of these initiated cells, during which time, an evolution of changes occurs in these initiated cells to bring about the malignant stage.

The objective of this research project is to develop a quantitative *in vitro* assay to detect chemicals which could be tumor promoters and to use this assay to study the mechanism of tumor promotion. The *in vitro* assay being used is based on measuring intercellular communication (i.e., "metabolic cooperation") between two strains of Chinese hamster cells. Validation of the assay, as well as the study of how intercellular communication might play a role in tumor promotion, has been under investigation.

To date, the Principal Investigator's results have been promising in that, using his *in vitro* assay, he has shown that he can eliminate confusion in the literature concerning the discrepancies between the results of the Ames *in vitro* assay for screening mutagens and *in vivo* results testing these chemicals for "carcinogenicity". For example, dinitrobenzene is positive as a mutagen in the Ames assay, negative as a tumor initiator in animal test, positive in the intercellular communication assay as an inhibitor of intercellular communication and positive as a tumor promoter in the mouse skin.

In addition, the Principal Investigator has shown that dieldrin, aldrin, polychlorinated biphenyls, polybrominated biphenyls, teleocidin (a natural promoter), mirex, kepone, 2,4,5-T, and 2,4D all inhibit intercellular communication at non-cytotoxic concentrations. The assay not only has proven its ability to validate known tumor promoters, but also to *predict* that certain chemicals would be tumor promoters *in vivo* from *in vitro* results. It has been shown that 2,4,5,2',4',5'-HBB, but not 3,4,5,3',4',5',-HBB is both an inhibitor of intercellular communication *in vitro* and a tumor promoter of rat liver tumors *in vivo*.

Lastly, a conceptual linkage of cells which inhibit intercellular communication *in vitro* to teratogens and reproductive toxicants has been made.

Title:

Development and Applications of Biomembrane Toxicity Electrodes

Principal Investigators:
K. H. Mancy,
M. E. Myerhoff,
B. Chin,
M. Brabec

Institution:
University of Michigan

Project Period:
3 years

Budget:
(2 years) $104,097

EPA Project Number:
R 808063

The objective of this project is the development of simple, but effective, rapid techniques for toxicity measurements. The underlying principle of these new sensors is the amperometric or potentiometric detection of changes in the biochemical activity of bacteria, mammalian cells, organelles or certain enzymes, affected by a toxicant or group of toxicants. Generally classified as biomembrane electrodes, these sensor systems may incorporate the biological material in an immobilized layer directly mounted on an ultrasensitive base electrode. The scope of this study includes the following sensor systems:

(1) Mutagenicity Sensor: Research findings indicate the feasibility of a mutagenicity sensor based on the principle of the Ames Assay. In this case, immobilized mutant strains of *Salmonella typhyrium* are mounted on an ultrasensitive oxygen electrode and incubated in the absence of histidine, which is required for growth. Respiratory activities were determined in the absence and presence of suspected mutagenic agents. In case of mutagenic transformation to histidine independence, growth of revertant bacteria is easily and rapidly sensed by an increase in respiratory activity. This procedure took no more than 3-4 hours, instead of the 2-3 days required to conduct the traditional Ames Assay.

An alternate test procedure will utilize *B. subtilis* M45 (Rec$^-$) which is genetically deficient in DNA recombination and *B. subtilis* H17 (Rec$^+$), the wild type. Both strains will be immobilized on two separate oxygen electrodes. Lethal mutation will kill the Rec$^-$ bacteria and will have less effect on the respiratory activity of the wild type Rec$^+$.

(2) Broad Spectrum Toxicity Sensors: This includes biomembrane electrode systems in which the biological membrane is made of immobilized (a) yeast cells, (b) mammalian tissue, and (c) mitochondria. The inhibition or alterations in the activity of the biological membrane, induced by a toxicant, will be monitored by determining oxygen uptake or carbon dioxide production using appropriate base electrodes.

(3) Automated Toxicity Biomonitor: This is an on-line automated biotoxicity assay utilizing ammonia gas potentiometric electrode. The reagents used are ammonia gas producing biocatalysts which serve as biological indicators for the screening of toxic substances. This included the use of Urea-Urease System for heavy metal screening and Amino Acid Oxidase System for organic compound screening.

(4) Disposal Pesticide-Nerve Gas Sensor: This sensor system is based on the inhibition of the enzyme acetylcholinesterase induced by certain pesticides and nerve gases. The detection of the immobilized enzyme activity is done by measuring the rate of production of acetic acid according to the following reaction:

$$\text{acetycholine} + H_2O \text{ -------}> \text{choline} + \text{acetic acid}$$

A newly developed polymer membrane pH electrode was used to monitor the reaction. This new pH sensor is inexpensive, disposable and replaces the costly fragile glass pH electrodes.

Title:
Anticholinesterase Insecticide Induced Pancreatitis—Chronic Studies

Principal Investigator:
Robert Gooddale

Institution:
University of Minnesota

Project Period:
1 year

Budget:
(1 year) $75,038

EPA Project Number:
R 808516

Accidental exposure to the organo-phosphate insecticide Diasinion® is known to cause acute interstitial pancreatitis and death from cholinergic crisis in man, dog and other domestic animals. The maximum tolerated bolus I.V. dose in dog is about 75 mg/kg in previous studies which were reported recently. The purpose of the present project is to see if chronic exposure to lower doses which do not cause overt symptoms or signs is harmful to the pancreas. In dogs, cats, rabbits, and guinea pigs, graded doses from 1 to 25 mg. per kilo per day will be administered S.C. to establish the dose level which is tolerated without significant weight loss or development of obvious weakness.

Then chronic doses at that level will be given up to 12 weeks. Small mini-infusion pumps will be implanted aseptically subcutaneously for constant rate of administration of agent. Venous blood will be drawn periodically and the level of serum cholinesterase depression will be measured. Serum amylase and lipase levels will also be measured periodically. Pancreata will be removed at the end of the experiment for light and electron microscopy. Focal acinar hyperplasia, neoplasia and pancreatitis will be specifically looked for. Suitable controls, utilizing infusions of solvent without Diazinon in normal animals will also be used.

There is mounting experimental evidence that excessive cholecystokinin and gastrin hormone levels stimulate pancreatic growth and even cause neoplasia in some experimental animals. It is possible that chronic cholinergic stimulation induced by organophosphate insecticide may also cause pancreatic hyperplasia or neoplasia or chronic pancreatitis. These possibilities will be investigated under the conditions described above.

Title:

Competitive Adsorption of Trace Organics from Natural Waters by Activated Alumina

Principal Investigator:
Mriganka M. Ghosh

Institution:
University of
Missouri-Columbia

Project Period:
2 years

Budget:
(2 years) $117,136

EPA Project Number:
R 809425

Many public water supplies in the U.S. contain arsenic and selenium in excess of maximum contaminant levels (MCL) established in the Safe Drinking Water Act. Many of these water supplies are located in small communities lacking elaborate facilities. Activated alumina seems to offer the most inexpensive method for the removal of these toxic metals. The objective of the proposed research is to investigate the specific adsorption of arsenic and selenium under competitive conditions, using both batch slurry reactors and continuous flow columns. The effect of such parameters as pH, ionic strength, interfering ions such as F^-, $SO_4^=$, Cl, $HCO_3^=$, Ca^{++}, Mg^{++}, Fe^{++}, SiO_2 and natural organics on the charge density and adsorption density of activated alumina will be studied. Various regenerant chemicals such as HCl, H_2SO_4, or NH_3 will be studied in an effort to develop an optimum regeneration scheme for alumina.

The project will begin with a literature survey of data on arsenic and selenium in U.S. waters, with particular emphasis on their speciation. Laboratory experiments on equilibrium adsorption will be conducted using alpha and gamma-Al_2O_3. Initial studies will involve single solutes of either As(V) or Se(IV). Data will be collected to determine kinetics of adsorption at different pH levels. In each case, the charge density and pH_{zpc} of alumina will be determined using alkalimetry. In next series of equilibrium studies, mixtures of metals will be used to determine competitive adsorption. Selected interfering ions will be tested in single metal adsorption experiments to determine their inhibitory effects, if any.

Phase two of the project will be devoted to column studies of adsorption of As and Se on alumina. Standard models, such as homogeneous surface diffusion model (HSDM) will be used to predict solute breakthrough profiles. Bisolute column runs will reveal any "chromatographic shift" in breakthrough profiles caused by competitive adsorption. In phase three of the project, experiments on the regeneration of spent alumina will be conducted using as variables such items as required concentration and bed volumes of caustic regenerant, and of acid rinse. Percent recovery of sorptive capacity, elution loss and change in surface properties of alumina will be monitored.

Title:
Transmission Electron Microscope Studies of Lichens Exposed to Air Pollutants

Principal Investigator:
Sharon Eversman

Institution:
Montana State
University-Bozeman

Project Period:
1 year

Budget:
(1 year) $23,457

EPA Project Number:
R 808932

Objectives:

(1) Complete a series of transmission electron microscope investigations of effects of four pollutant conditions: SO_2, O_3, SO_2/O_3 combination, PAN.
(2) Correlate ultrastructural observations with recorded pollutant effects on metabolism of six lichen species.
(3) Determine when ultrastructural repair occurs in light/dark conditions.
(4) Observe synergistic effects that might occur with SO_2/O_3 combinations.
(5) Compare lichen responses with responses observed in two grass species.

Methodology

(1) Two lichens, *Hypogymnia enteromopha* and *Parmelia sulcata*, were fumigated with PAN in chambers at the Air Pollution Research Center, University of California-Riverside. Photosynthesis rates and distributional records were determined: ^{14}C method was used for photosynthesis rates.
(2) Two lichens, *Parmelia caperata* and *Ubilicaria muhlenbergii*, were fumigated with SO_2, O_3, and SO_2/O_3 fumigations at various dosages, combinations, and time schedules followed by different light and dark periods in growth chambers at Oak Ridge National Laboratory. Photosynthetic rates were determined by a ^{14}C method.
(3) Two lichens, *Usnea hirta* and *Parmkelia chlorochroa*, were fumigated with SO_2 in field conditions. Photosynthesis rates (infrared gas analyzer), respiration rates (manometer), plasmolysis rates of algal cells (light microscopy) and external thallus conditions were recorded.
(4) Two grasses, *Agropyron smithii* and *Koeleria cristata*, were collected from the same SO_2 field fumigation system as the two preceding lichen samples. Leaf conditions and physiological responses for the grasses were recorded.
(5) *Usnea hirta* and *Parmelia chlorochroa* lichen specimens were collected 1-2 km from two 350-megawatt coal-fired power plants in Colstrip, Montana in 1979.
(6) All specimens were fixed and embedded immediately after collection or upon receipt of specimens after treatment. Standard procedures for electron microscope preparation were followed: fixation in 3% glutaraldehyde and 1% osmium tetroxide, dehydration in acetone series, and embedding Spurr's resin.
(7) Quantitative estimates of ultrastructural changes are made by counting numbers of plastoglobuli and pyrenoglobuli in chloroplasts, measuring sizes of those same lipid bodies, determining distances between thylakoid membranes, counting and measuring sizes of starch grains in the chloroplasts. These measurements are made on electromicrographs.

Title:
Habitat Selection in Benthic Marine Invertebrates Relevant to Community Patterns Near Waste Discharges

Principal Investigator:
J. Nybakken

Institution:
Moss Landing
Marine Laboratories

Project Period:
6 months

Budget:
(2 years) $143,377

EPA Project Number:
R 808782

There is a general belief that benthic community changes near marine waste discharges are related to pollutant tolerance. The most worrisome pollutants are heavy metals and certain hydrocarbons. However, the individual and synergistic effects of these pollutants are confounded by the effects of organic enrichment. Unlike these chemical pollutants, properly managed coastal enrichment may have positive environmental consequences.

A series of complementary field and laboratory experiments to distinguish the community effects of organic enrichment from heavy metal and hydrocarbon pollutants will be undertaken. These experiments primarily concern behavioral attraction or avoidance to a modified sedimentary habitat.

This study concerns habitat selection rather than survival rates, which is the subject of most bioassay studies. Many animals may make choices that determine community patterns before differential survival is relevant. Once an individual settles, survival is obviously critical.

The experiments focus on species that represent the three major components of community change around waste discharges: (1) a group of "weedy" species at the disturbance center, (2) a group from the native assemblage with elevated abundance at the disturbance edge, and (3) the native group of species outside the zones of gross community change.

It is hypothesized that the pollutant resistant concept is not generally correct and that community patterns near waste discharges are primarily linked to enrichment and habitat selection. Similar patterns and processes may characterize naturally enriched benthic communities as well.

Title:

Oxidation-Reduction Behavior of Chlorinated Dioxins on Particulate Matter

Principal Investigator:
G. A. Eiceman

Institution:
New Mexico
State University

Project Period:
2 years

Budget:
(1 year) $34,431

EPA Project Number:
R 809084

Polychlorinated dibenzo-p-dioxins (PCDD) are a series of tricyclic aromatic compounds containing one to eight chlorine atoms. Certain isomers of these chlorinated dioxins have been associated with several threats to animal and human health and the environment. While well-recognized sources of chlorinated dioxins in the environment are industrial wastes and release of certain herbicides, chlorinated dioxins may also be released through combustion processes including incineration of municipal refuse. Although the origin of PCDD in fly ash from municipal incinerators is uncertain, Towsend's theory has been proposed to account for the distribution of isomer ratios in particulate matter which has been released into the atmosphere for municipal and other incinerators.

The theory was developed from Townsend's observations that distribution ratios of chlorinated dioxins (i.e., tetra to penta, penta to hexa) were uniform among environmental samples for a wide range of PCDD concentrations. An oxidation-reduction model was proposed to account for these results including reduced levels of lower chlorinated dioxins in particulate matter which had been exposed to atmospheric oxidation. Townsend also showed that conditions of incineration, not feedstock, seemed to determine relative ratios of PCDD at the point of source. The practical implications for pollution abatement of PCDD are enormous if these theories are correct. For example, changes in incineration parameters will not eliminate the formation of T_4CDD, but only control the absolute amounts. Alternately, if chlorinated dioxins actually undergo oxidation processes on particulate matter, the addition of excess air with hot particulate matter may greatly reduce the concentration of all PCDD in incinerator emissions.

Information and answers will be sought for the following questions:

(1) Are PCDD oxidized between temperatures of 60 and 300°C on particulate matter (in this case, the finest sieve fraction of fly ash from electrostatic precipitators)?
(2) What are the products of the oxidation processs:
(3) Do certain conditions promote oxidative destruction of PCDD more than others and what are these conditions?
(4) Are PCDD reduced or chlorinated under certain conditions?
(5) Do values for chlorinated dioxin ratios quantitatively agree with results from the studies of Townsend?

Research in this project includes the use of a reaction chamber for modeling the behavior of PCDD on fly ash. Conditions of this reaction chamber may be closely controlled to simulate conditions of emission plumes for this study. Conditions such as gas composition, particulate temperature, residence times for dioxins, and other will be varied. Experimentally, known amounts of dioxins are released onto fly ash at selected temperatures for certain lengths of time. The reaction is quenched and products from the treatment process are analyzed using analytical methods based on high resolution gas chromatography (GC) and gas chromatography/mass spectrometry (GC/MS). Initially, T_4CDD and OCDD will be used with atmospheric conditions using several air-helium ratios. After reactions using oxidative conditions are characterized, results from using several HCl/helium and HCl/air/helium mixtures will be examined.

Title:

Enzyme Electrode Probes for Organohalogen Compounds

Principal Investigator:
George G. Guilbault

Institution:
University of
New Orleans

Project Period:
2 years

Budget:
(1 year) $51,774

EPA Project Number:
R 808532

An immobilized enzyme electrode, specific for halomethanes (chloroform, bromoform, dichloromethane, etc.) will be developed, using an enzyme induced from a selected microbial species, purified by electrophoresis, DEAE and affinity chromatography, and chemically bound directly onto a chloride or silver electrode. A hydrophobic Celgard or Teflon-membrane will be used to separate the enzyme layer from the media, thus preventing halides (Cl^-, Br^-, F^-) from penetrating the outer membrane. Only the small halomethanes will enter, be acted upon by the immobilized enzyme layer to yield Cl^- or Br^-, which will be detected by the base Cl^- (Ag) electrode.

Investigations done by many researchers have revealed that numerous microorganisms such as Red Algae, Fungi *Fusarium oxysporum, Aerobacter aerogenes,* etc., are capable of transforming organochlorine compounds through metabolic pathways.

The microbial species *Pseudomonas putida* has been selected. Its growth has been optimized by using a liquid media made up of certain proportions of essential chemicals and nutrient solutions. The chloroform concentration has been increased in subsequent growth trials. A number of preliminary assays on whole cell bacteria have been carried out to confirm that the selected induced bacteria are quite capable of degrading the halomethanes. NBS microferm fermentor was used to optimize mass culture of the bacteria in large quantities by means of culture jar.

The bacteria are being studied in order to proceed to the fractionation and separation, isolation and purification, and finally immobilization of the enzyme for the degradation of these halomethanes.

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Title:
Comutagens in the Environment

Principal Investigator:
Toby G. Rossman

Institution:
New York University
Medical Center

Project Period:
2 years

Budget:
(2 years) $190,304

EPA Project Number:
R 808482

Experimental and epidemiological data suggest that some agents can contribute to the development of human cancer in subjects which have been exposed to very low levels of carcinogens. In some cases, the agent in question is not a mutagen in any standard mutagenesis bioassay.

Preliminary data is presented showing that two substances produced as a result of fossil fuel combustion, arsenite and SO_2, act as comutagens (enhancers of mutagenesis). Neither agent is mutagenic itself under conditions used in these experiments, in which there is low toxicity. The comutagenic effect of SO_2 is seen in both prokaryotic (*E. coli*) and eukaryotic (Chinese hamster V79) cells. Both agents enhance UV-induced mutagenesis in *E. coli* by a factor of eight- to ten-fold.

The mechanisms of action of SO_2 and arsenite comutagenesis will be determined. Factors to be examined will include the role of free radicals, possible inhibition of DNA repair and the nature of mutagens with which these agents act as comutagens. The comutagenesis assay systems will be further developed along a number of lines, including the use of human cell lines (normal and xeroderma pimentosum), the use of bacterial mutants deficient in a variety of repair pathways, the use of a variety of chemical mutagens causing different types of DNA damage, and the inclusion of rat liver S9 to metabolize possible comutagens.

Agents to be examined as possible comutagens include pyrene, fluoranthene, nitrites, nitrates, various halogenated hydrocarbons, toluene, phenol and xylene. Some of these agents have already been identified as cocarcinogens.

Title:

Anaerobic Biodegradation of Toxic Aromatic Compounds

Principal Investigator:
Lily Y. Young

Institution:
New York University
Medical Center

Project Period:
3 years

Budget:
(2 years) $166,266

EPA Project Number:
R 809077

Hazardous aromatic hydrocarbons are principally destroyed in the environment by microbial action. The aerobic mechanism, though well understood, cannot account for the decomposition of these compounds under anaerobic conditions. The proposed research intends to examine the molecular fate of monocyclic aromatic hydrocarbons with the following tasks:

(1) Determination of the rate and extent of biodegradation by strictly anaerobic microbial consortia for a selection of compounds differing in ring substituent complexity.
(2) Isolation and characterization of the major species responsible for the metabolic transformations.
(3) Identification of the major intermediates formed during the catabolic sequence for each compound. Methodologies developed for this task include analyses by GC, HPLC, and GC/MS procedures.
(4) Development of a conceptual model for the anaerobic degradation of this class of compounds which could aid in the prediction of the fate of related toxic compounds.

Title:

Airborne Toxic Substances and Airway Epithelial Function

Principal Investigator:
R. Boucher

Institution:
University of North Carolina-Chapel Hill

Project Period:
2 years

Budget:
(2 years) $258,588

EPA Project Number:
R 808753

Effects of inhaled agents on airway epithelial functions may contribute to the pathophysiology of occupational lung diseases. We previously demonstrated active Na^+ absorption across human and canine bronchial epithelium, *in vitro*, which may provide a mechanism for the regulation of airway surface liquid volume. In the present experiments third to fifth generation canine bronchi were mounted in Ussing chambers and short circuited. Ion fluxes and bioelectric properties were monitored for 90 minutes before addition of formaldehyde (F). After 60 minutes of luminal exposure to 0.1, 0.3, 1.0 and 3.0 mM/L F, short circuit current was reduced by 12, 21, 60, and 90%, while conductances changed by -4, -14, -39 and -28%, respectively. These changes were poorly reversible and serosal exposure to 3.0 mM/L was without effect. Luminal exposure to 1.0 mM/L reduced Na^+ absorptive flux from 3.02 to 2.52 and increased backflux from 1.51 to 2.21 $ueq/cm^2 \cdot hr$. Both unidirectional Cl^- fluxes were reduced from about 5 to 3 $ueq/cm^2 \cdot hr$. ^{14}C-Mannitol permeability was unchanged. We suggest that the interaction of F with the apical membrane of epitheiial cells perturbed transcellular ion flows. Cl^- flows were diminished whereas passive Na^+ flux increased. Decreased $Na^=$ active flow implies an additional action on the Na^+ transport process. Preliminary results suggest a similar pattern of effects on human bronchial epithelium, *in vitro*. These effects could increase airways liquid in individuals exposed to formaldehyde vapors.

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Title:
Atmospheric Transformation and Bioactivity of Combustion from Residential Wood Stoves

Principal Investigators:
Richard M. Kamens,
Mark D. Sobsey

Institution:
University of North Carolina-Chapel Hill

Project Period:
3 years

Budget:
(2 years) $220,030

EPA Project Number:
R 808112

The objective of this research is to characterize the transformation processes and associated bioactivity of dilute combustion products from commercially available residential wood heaters in winter time atmospheres containing simulated O_3, HC and NO_x pollution. The relative importance of the already identified pathways for polyaromatic hydrocarbon destruction on soot particles (attack by light, O_3, and other smog products) will be assessed. The biological mutagenicity of freshly diluted wood emissions will also be compared to that of emissions which have had an opportunity to age and react within an outdoor chamber air parcel.

Combustion products from a commercially available woodstove will be injected and diluted into two outdoor smog chambers. This will permit the PAHs on soot particles to be exposed and transformed under natural winter conditions. One chamber will be a control, containing only wood emissions in unpolluted air. In the other, soot will be exposed to light and dark and ambient levels of O_3, NO_2 and dilute smog. Changes in 4-6 representative PAHs will be monitored over the course of 4-8 hour experiments. The Ames bioassay test and other mutagenicity tests will be used to measure the mutagenicity of the chamber soot particles.

The Principal Investigator expects: (1) to quantify the relative importance of light, O_3, and other radical sources on the transformation of wood combustion PAHs on adsorbed soot; (2) determine the extent of change in relative mutagenicity after reaction with light, O_3, NO_2, and dilute smog; and (3) produce a complete data set which will include aromatic PAH transformation, associated bioactivity, combustion conditions and energy release rates from typical residential wood burners and thus will be useful in evaluating the environmental and energy impacts of increased wood burning.

Title:
An In Situ *Analyzer for Organic and Elemental Carbon in Ambient Aerosols*

Principal Investigator:
James J. Huntzicker

Institution:
Oregon Graduate
Center for Study
and Research

Project Period:
2 years

Budget:
(2 years) $120,000

EPA Project Number:
R 808087

An *in situ* instrument for the analysis of organic carbon and elemental carbon in ambient aerosols will be developed. This instrument will be a modified version of the Oregon Graduate Center laboratory organic and elemental carbon analyzer. Fine particle aerosol will be sampled through a virtual impactor head or other size discriminator and collected on a glass fiber filter located in the combustion zone of the carbon analyzer. At the end of the 30-minute sampling period the organic and elemental carbon concentrations will be measured by a combustion process which involves volatilization of organic carbon at 600°C in a helium atmosphere followed by combustion of elemental carbon at 600°C in a 2% oxygen/98% helium atmosphere. All evolved carbon will be oxidized to CO_2, reduced to CH_4, and measured in a flame ionization detector. The transmission of 633 nm He-Ne laser light through the filter will be monitored continuously throughout the sampling and analysis. The transmission data are used to correct for the pyrolytic conversion of organic to elemental carbon which occurs during the organic analysis. The complete sampling and analytical cycle requires about one hour.

The instrument will be tested in both the laboratory and the field for analytical efficiency, organic artifacts, and particle size effects. The first field test of the prototype instrument will take place during the EPA Denver Haze Study in January 1982. More detailed field testing will be conducted in Portland, Oregon and Pasadena, California. The instrument will be operated in the field in conjunction with other air quality monitors (e.g., nephelometer, ozone monitor, etc.) and the data used to investigate the question of secondary organic aerosols (i.e., organic aerosols formed as a result of atmospheric chemical reactions).

It is anticipated that the hourly organic and elemental carbon data coupled with meteorological and source emission data will permit a more accurate assessment of the importance of secondary organic aerosols than previously possible.

Title:
Laboratory Investigation of Buoyant Emission Particulate Matter

Principal Investigator:
James W. Deardorff

Institution:
Oregon State University

Project Period:
2 years

Budget:
(2 years) $138,256

EPA Project Number:
R 808919

In this research, the dispersion of buoyant stack effluents into the daytime mixed layer over simple terrain is modeled, using a convectively mixed layer in the laboratory as the modeling facility. The method has previously been applied successfully to model dispersion of non-buoyant stack effluents. The main effect of a mean wind, U, is simply modeled by replacing pollutant travel time with downstream distance divided by U.

The technique is to utilize fluorescent dye emitted from a towed stack and optically activated by a laser beam directed along the cross-plume direction. When not illuminated, dyed fluid along the optical line of sight to the beam is invisible and does not greatly attenuate the signal, which is linear with concentration over three decades of intensity.

Mixed-layer scaling will be applied to all quantities to determine how the (dimensionless) mean ground-level concentration depends upon downstream distance and mean wind, stack-effluent buoyancy, and initial angle of bending-over of the plume. The actual stack height will be modeled as 1/10 the mixed-layer height. A characteristic value for the dimensionless mixed-layer capping-inversion strength will be modeled. The emphasis will be on determining mean maximum ground-level concentration and how it depends on the meteorological and stack-effluent variables.

Results should prove very useful in interpreting field measurements downstream of a tall stack in the daytime even over complex terrain. Guidance is needed on what the expected concentration distribution would be over simple terrain under a given set of meteorological stack-effluent conditions so that effects of complex terrain, in the daytime, can be distinguished from effects of convective mixing and effluent buoyancy.

Title:

Effects of Pentavalent Arsenicals on Resistance of Mice to Salmonella *Infection*

Principal Investigator:
Frank W. Judd,
Norman L. Savage

Institution:
Pan American University

Project Period:
2 years

Budget:
(2 years) $104,164

EPA Project Number:
R 807843

Arsenical compounds, especially pesticides, are widely and frequently indiscriminantly used. Non-target organisms including man, and wild and domestic mammals are subject to contamination by these compounds. Many of the more subtle biological effects of these compounds on non-target organisms are poorly understood, particularly any alterations in the host's infectious disease susceptibility and ability to resolve an infectious disease. Information has been published on viral disease susceptibility, but no information is available on bacterial disease susceptibility. Since some arsenicals have been used in treatment and prevention of certain bacterial diseases and it is known that arsenicals may be toxic to soil bacteria as well as to animals, it is possible that synergistic effects between microorganisms, host and arsenicals may affect the process of infection and subsequent disease progression in animals exposed to arsenical compounds.

The principal objective of this project is to evaluate the effects of selected pentavalent arsenical compounds on the resistance of albino mice to infection with *Salmonella typhimurium*. Subordinate objectives include a determination of the importance of variation in concentration of the arsenicals, time relationships of challenge with arsenicals and *Salmonella*, and whether the exposure is acute or chronic. Also, the mechanism(s) of action of the arsenicals on the disease process will be investigated.

Four pentavalent arsenicals will be examined (monosodium methanearsenate, cacodylic acid, sodium arsenate and arsanilic acid). *Salmonella typhimurium* will be utilized as the infecting agent. Female, ICR Swiss mice will be used as subjects. The effects of arsenicals administered both acutely by stomach intubation and chronically in the drinking water at times before, simultaneous with, and after *Salmonella* i.v. inoculation will be compared (based on the mortality rate and total deaths over 30 days following inoculation) with the development of the disease administered orally (normal route). Controls will consist of animals receiving water only, *Salmonella* only, and an arsenical only.

Those compounds and procedures producing significant alterations in the progression of the salmonellosis will be subjected to investigations to identify the mechanism(s) of action of the arsenicals. Tissue levels of arsenic will be determined and used *in vitro* with the bacterium to ascertain if these concentrations are toxic to the bacterium. Tissue concentrations of baceria will be determined shortly after infection so as to evaluate the effectiveness of the initial phagocytic capabilities of the mice. Tissue concentrations of bacteria will be determined during the course of the disease as an indicator of the ability of the mice to develop an immune response to resolve the infection.

The potential alteration of macrophage function in arsenic treated animals will be examined. The development of the immune status of mice receiving arsenicals will be determined by immunizing groups of animals with killed vaccines (humoral response) or sublethal doses of live cells (cell-mediated response). Humoral protection is expected to be manifest as a rapid clearance of bacteria from the blood. Cell-mediated protection is expected to result in a rapid clearance of organisms from the spleen and liver. Overall protection should result in reduced mortality.

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Title:

Diagnosis and Prediction of Atmospheric Transport on Regional and Synoptic Scales Using a Three-Dimensional Dynamic Model

Principal Investigator:
Thomas Warner

Institution:
Pennsylvania State University

Project Period:
2 years

Budget:
(2 years) $149,971

EPA Project Number:
R 808610

The various facets of the proposed research all have the common goal of providing a procedure whereby a three-dimensional (3-D) dynamic model may be used to provide meteorological data for atmospheric transport calculations on the regional and synoptic scales. The demand for improved transport calculations arises because of the pressing scientific, legal, and environmental problems that require a knowledge of the path followed by pollution-laden air parcels as they move from source to receptor. However, no general procedure is available for calculations of parcel trajectories that demonstrates accuracy over a broad range of conditions.

Many techniques exist, but most have limitations such as being site-specific, being applicable under only certain meteorological conditions or times of day, relying on the synoptic-scale rawinsonde network for wind data or being applicable to only specific levels in the atmosphere. A procedure that might happen to be relatively successful in some situations provides very poor trajectory estimates in other situations. It is difficult to determine *a priori* which procedure will be best in a given case. Thus, the need for a general and reliable technique for atmospheric transport computations is evident.

In this program the Penn State 3-D mesoscale dynamic model will be adapted so that it can be used to provide atmospheric data on the regional or synoptic scale for short-, medium-, or long-range transport calculations in the planetary boundary layer or the free atmosphere, including the upper troposphere and lower stratosphere. It will be applicable under a wide range of meteorological conditions as well as surface conditions that are characterized by complex terrain and complicated land use patterns. After verifying model-predicted trajectories against existing tetroon data, the model will be applied to a transport problem involving complex terrain under stable conditions over the eastern United States.

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Title:

Investigation of Polymerization of Xenobiotic Aromatic Compounds (e.g., Some Pesticides) and Their Incorporation into Soil and Sediment Humus

Principal Investigator:
J-M Bollag

Institution:
Pennsylvania State University

Project Period:
3 years

Budget:
(1 year) $69,022

EPA Project Number:
R 808165

With the increasing use of pesticides and xenobiotics in nature there has been a growing concern with regard to the fate of these substances and their unintentional effects within the environment. Within the last few years special consideration has been given to the 'bond residue' problem, the formation of molecules far more complex than the parent molecule. The occurrence of this latter phenomenon in the soil environment, where specific binding to organic matter to form highly esoteric materials can occur, is of concern to us.

Naturally occurring aromatic compounds are often complexed or polymerized during humus formation by the activity of phenoloxidases and other catalysts from soil, but little is known about the activity of these catalysts on aromatic xenobiotics. Therefore, it appears of great interest to focus attention on the effect of the "polymerizing" enzymes and other soil catalysts on the transformation of various pollutants. One of the goals of this research is to isolate and identify products of oxidative coupling reactions of various pesticide intermediates and other xenobiotics (halogenated phenols, various naphthols, substituted anilines) with naturally occurring phenolic soil constituents. Until now the polymerizing activity was established under typical laboratory conditions but now this observation should be confirmed in natural environments. It is also planned to develop an assay in order to determine the polymerizing potential of soils and sediments. Such an assay could be very useful in screening various environments for their possible 'binding' effect on xenobiotics.

Polymerization or condensation reactions involving xenobiotics may lead to the formation of new chemicals whose impact on the environment is unknown. Even if these xenobiotics are incorporated into natural complexes such as humus or sediments, they present a source of concern as probable environmental pollutants, since their later release from such complexes is quite possible.

Title:
Oxidation of Alkoxy Radicals

Principal Investigator:
Julian Heicklen

Institution:
Pennsylvania State University

Project Period:
2 years

Budget:
(2 years) $159,893

EPA Project Number:
R 808528

Alkoxy radicals are produced in the atmosphere from the oxidation of hydrocarbons. They can be removed by oxidation, decomposition, or isomerization. The Principal Investigator proposes to investigate the competition between the various reactions paths to determine the appropriate rate coefficients as a function of pressure and temperature from 25-150°C. Also for those alkoxy radicals that isomerize, the products and mechanisms of the removal of the product radicals will be investigated. In addition the rate coefficient for CH_3O oxidation will be obtained by competition with the $CH_3O + NO_2$ reaction, since CH_3O neither decomposes nor isomerizes below 150°C.

The radicals will be generated by the photolysis of the corresponding alkyl nitrates in the presence of NO and O_2. Products of the reaction will be measured by gas chromatography. The pertinent reactions are:

$$RONO + H \rightarrow RO + NO \qquad \text{Rate} = \Phi I_a$$

$$RO + NO \rightarrow RONO \text{ (or } R'O + HNO) \qquad 1$$

$$RO + O_2 \rightarrow R'O + HO_2 \qquad 2$$

$$RO \rightarrow \text{not } R'O \qquad 3$$

where RONO is the alkyl nitrite and *R'O* is the corresponding aldehyde. This mechanism leads to the rate law

$$\Phi[R'O]^{-1} = (1 + k_1 [NO]/k_2 [O_2] + k_3/k_2 [O_2])$$

so that plots of $\Phi[R'O]^{-1}$ vs $[NO]/[O_2]$ at constant $[O_2]$ or $[O_2]^{-1}$ at constant [NO] will give straight lines whose slope to intercept ratios will give, respectively, k_1/k_2 and k_3/k_2.

Title:

Kinetics of Aromatic Combustion and Polycyclic Aromatic Hydrocarbon Formation in Liquid Fuels

Principal Investigator:
Normand M. Laurendeau

Institution:
Purdue University

Project Period:
2 years

Budget:
(2 years) $160,646

EPA Project Number:
R 808672

New liquid fuels derived from coal or shale oil will have high aromatic contents compared to petroleum-based fuels. Little information is available on the combustion kinetics of aromatics. As fuel aromaticity increases, formation of polycyclic aromatic hydrocarbons (PAH) and soot becomes more likely. PAH compounds may also be precursors of soot formation.

The objective of the research is to determine reaction rates and to identify reaction mechanisms for (1) the combustion of aromatic fuels, and (2) the formation and destruction of polycyclic aromatic hydrocarbons (PAHs). In this, the first phase of an anticipated multiphase effort, we will investigate the combustion kinetics of benzene. Special attention will be given to the influence of stoichiometry and temperature on combustion rates, reaction mechanisms, PAH formation, PAH destruction and soot formation.

Small quantities of benzene will be added to a subatmospheric (0.1 atm) hydrogen-oxygen flame stabilized on a flat flame burner. In this way, the well-known hydrogen-oxygen kinetics can be used to isolate the effects of aromatic combustion. Expected ranges of temperature and stoichiometry are $1200 < T < 1800$ K and $0.5 < \Phi < 2.0$, respectively. Concentration profiles will be measured using a water-cooled quartz microprobe and gas chromatography. Radical profiles, such as OH, will be measured via UV absorption and fluorescence spectroscopy. Measurement of selected PAH profiles will be attempted by two methods: (1) glass capillary gas chromatography/mass spectrometry, and (2) laser-induced fluorescence spectroscopy. Reaction mechanisms will be identified by comparing measured and predicted rates of concentration profiles, especially those of carbon-containing species.

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Title:

Mechanisms of Mutagenesis by Environmental Carcinogens

Principal Investigator:
Barbara H. Rosenberg

Institution:
Sloan-Kettering Institute for Cancer Research

Project Period:
3 years

Budget:
(2 years) $215,157

EPA Project Number:
R 809550

The major goal is to identify specific pathways of genetic change which result from specific types of chemically-induced lesions in DNA and which play an important role in carcinogenesis.

The principal cellular target of environmental carcinogens is generally assumed to be DNA, and the mechanism of carcinogenesis is thought to involve mutational events during DNA replication and/or repair synthesis. However, although it is known that chemical carcinogens produce many types of lesions in DNA, the relationship between these lesions and mutagenesis has not been proven and the molecular events that may lead from the responsible lesions to permanent genetic change are unknown. Moreover, it has not yet been established which DNA lesions produced by a particular carcinogen and which types of genetic change may be irrelevant and which may be responsible for the production of cancer. These questions are addressed in this project, which is designed to study the molecular basis of mutagenesis by environmental carcinogens.

We propose to correlate the carcinogenic potentials of various chemicals and the types of DNA adducts (or other lesions) they form with the extent and nature of cellular repair of the lesions and with the kinds of DNA replication defects that may occur before or after repair.

We will use the mammalian virus SV40, with its DNA modified *in vitro* by treatment with the carcinogen. The modified and characterized SV40 DNA vector will then be introduced into mammalian cells, where its replication will be studied under conditions in which no transcription of the modified DNA is necessary.

Title:
Mutagenic DNA Modification

Principal Investigators:
Gerhard Stohrer,
Tsanyen Yang

Institution:
Sloan-Kettering
Institute for Cancer
Research

Project Period:
3 years

Budget:
(2 years) $136,929

EPA Project Number:
R 808746

The aim of this research is to study *mutation* by *environmental carcinogens* at the molecular level. *Adducts* of carcinogens with nucleotides will be introduced into a specific position of a phage genome to study the mutagenic consequences of such a defined lesion. The *site-specific mutagenesis* system of Bhanot, Khan and Chambers will be adapted to the study in the following way:

(1) A primer will be synthesized and carcinogen adducts will be introduced into the primer by the solid state phosphotriester method of Gait *et al.* An alternative will be the enzymatic introduction by either *E. coli* polymerase I or terminal deoxynucleotide transferase. Our preliminary experiments with modified deoxynucleotidetriphosphates suggest that the enzymatic route may be feasible. The adducts will initially include those of guanine with *pyrenyl oxirane* (an analog of the environmental carcinogen benzo(a)pyrenediolepoxide) and those of thyumine and guanine with *acetoxyxanthine*.

(2) In the enzymatic approach, a second primer, immediately following the modified position will be synthesized. It will be ligated to the first primer carrying the lesion by DNA ligase and the new prime, 1 + 2 purified.

(3) The viral strand of Φ *174* am-3 will be used as template and the am-3 site will by hybridized and saturated with restriction fragment Z-7 prepared from wild-type DNA. This is designed largely as a means to eliminate the progeny of the template strand.

(4) Closed duplexes will be separated from remaining viral strands by nitrocellulose rather than nuclease S_1.

To complement this site-specific mutagenesis system, a traditional mutagenesis system based on *in vitro* treatment of replicative form Φ *DNA* with activated carcinogens and *transfection of spheroplasts is being developed* in this lab. This system will study reversions and frame shift mutations at the same locus as that used for the site-specific system. While the lesions will be characterized in the site-specific system, they will contain the whole mixture of lesions produced in the traditional way of mutagenizing DNA, some with known structures, others yet unknown. The two systems of mutagenesis outlined will together recognize the most relevant lesions involved in mutagenesis and carcinogenesis.

Title:
Atmospheric Chemistry of Natural Hydrocarbons

Principal Investigator:
Richard Kenley

Institution:
SRI International

Project Period:
2 years

Budget:
(2 years) $139,690

EPA Project Number:
R 808110

This work is designed to help evaluate the relative importance of urban and industrial hydrocarbons compared to natural hydrocarbons in ozone development in parcels of polluted air undergoing transport. The reaction pathways and products of the natural hydrocarbon-O_3 system will be elucidated in laboratory studies. Dilute mixtures of O_3, (0.1-1 ppm) and the hydrocarbon isoprene (1-10 ppm), or one of four terpenes to be studied (-pinene, -pinene, limonene, and myrcene) will be mixed in a glass reactor at 1 atm total pressure. The decrease in hydrocarbon will be monitored by gas chromatography. The low vapor pressure products will be used to enhance the sensitivity of the method. The OH-radical levels present will be determined by isobutane (or cyclohexane) addition.

Two methods will be used to study the OH-radical reaction of the same hydrocarbons: (1) a low pressure flow system, and (2) a solar induced reaction at atmospheric pressure (5 liter bulb). In the flow system, hydrogen atoms will be generated in a flow discharge at NO_2 added downstream to generate OH-radicals: $H + NO_2 + OH + NO$, HPLC, GC/MS, field ionization mass spectroscopy, and FT-NMR will be employed to identify products. The products of the reactants studies at high pressure will be identified by GC or LC techniques following extraction with a suitable solvent.

Title:
Generation Chemistry and Reactions of Chlorine Dioxide for Water Treatment

Principal Investigator:
Paul V. Roberts

Institution:
Stanford University

Project Period:
2 years

Budget:
(2 years) $144,414

EPA Project Number:
R 808686

The use of chlorine dioxide for the disinfection of potable water supplies offers some substantial advantages compared to chlorine. However, additional research on the chemistry of chlorine dioxide is needed to improve the economy of generating pure chlorine dioxide for water treatment. This research will identify experimentally the optimum reaction conditions for the liquid-liquid and the gas-liquid chlorine-chlorite reactions. This information is a prerequisite for designing a chlorine dioxide generation system utilizing either of these reactions.

This research will also promote understanding of the behavior of chlorine dioxide added to representative water supplies and will identify and quantify the inorganic end products of chlorine dioxide. These data are required to complement the results of toxicological studies of chlorine dioxide and its inorganic by-products so that the health effects of chlorine dioxide can be intelligently assessed.

A possible application of this research is the development of a chlorine dioxide generation process suitable for water treatment which achieves a high yield of virtually pure chlorine dioxide from chlorine and sodium chlorite, without significant amounts of either excess chlorine, unreacted chlorite, or undesirable chlorate byproduct being present in the reactor product.

The specific objectives are:

(1) To evaluate the available information concerning the chemistry of chlorine dioxide generation by the chlorine-chlorite process, and the behavior of the final product in aqueous solution.

(2) To determine the stoichiometry and yield of the chlorine-chlorite reaction in aqueous solution as a function of:

  a. the absolute concentrations of the reactants chlorite and chlorine
  b. the ratios of reactant concentrations
  c. the speciation of the reactant chlorine: Cl_2, HOCl, and OCl^-.

(3) To determine the stoichiometry, kinetics, and yield for two-phase gaseous chlorine-aqueous chlorite reaction as a function of:

  a. the absolute concentration of aqueous phase chlorite
  b. the mole fraction chlorine in the gaseous phase
  c. the ratio of reactants chlorine and chlorite in the reaction zone
  d. the pH of the liquid phase.

(4) To determine the range of concentrations of chlorine species to be expected in water supplies treated with chlorine dioxide generated by the chlorine-chlorite reaction.

Title:

The Nitella *Meadow in Clear, Oligotrophic Lakes: Function and Vulnerability to Shading*

Principal Investigator:
Raymond G. Stross

Institution:
State University of New York at Albany

Project Period:
3 years

Budget:
(2 years) $47,792

EPA Project Number:
R 808537

The primary goal of the research is to discover the mechanism of depth zonation in the deep dwelling charophyte, *Nitella flexalis* (L) Ag. Preliminary study has shown that density of shoots and boundaries of the meadow may be determined by separate factors of the environment. The first hypothesis to be developed is that light, acting through a photomorphogenetic receptor, may determine the lower boundary. The hypothesis is cast for both field and laboratory investigation.

Underwater spotlights, equipped with colored filter have been installed in Lake George, New York, where a vast underwater meadow of *Nitella* with sharp lower boundaries exists. The light beams are trained on the lake bottom immediately below the lower boundary of the meadow. They are intended to supplement ambient light in specific wavebands. Five treatments without replication are contained in the first series: red, green, blue, blue plus red, and a white light control. Success will be determined by germination and subsequent development of shoots of *Nitella*, within the beams of light. Both oospores and vegetative shoots will be deposited in special containers within the beams to serve as a source of propagules. Since the sediments in the study area are covered with a fishmoss, *Dicotomosiphon tuberosus*, the propagules of *Nitella* will be tested both in contact with and isolated from the fishmoss. Evaluation will be conducted from photographs taken of the test sites with the aid of SCUBA at periodic intervals. An additional control will consist of transporting a section of the *Nitella* meadow to locations within and outside of the light beams for the purpose of measuring rates of elongation relative to those inside the meadow. The rate of recolonization within the meadow is being measured. Permanent quandrats have been installed and the shoots have been removed from one-half of each quandrat.

The basis for a photomorphogenetic switch mechanism as a determinant of the lower boundary is also being investigated in the laboratory with oospores and later with the nodal-cell propagules. The question being asked, is light essential to activation of germination and is light necessary for differentiation of the larval plant (protonema) into shoot and rhizoid?

Shoots containing ripening oospores were transferred from the *Nitella* meadow into Lake George to dim light (1-3μ Ein x μ^{-2}) at July photophases and at 15°C. After falling into the substratum the oospores were harvested and set into a series of experiments designed to determine the effect of stratification, photoperiod, substratum, and organic extract on the capacity to germinate. If protracted incubation at low temperature and in the dark will render the oospore ready for activation, the follow-up experiments will be to determine whether light is essential for both activation and later differentiation of the germling. Fluence rate, wavelength and duration of light will be the primary variables. The requirements for development of ooospore and vegetative nodal cells will be compared.

Several other objectives are underway or are anticipated. The second objective is to describe the annual cycle of *Nitella* in Lake George and to simulate it in the laboratory, including fruiting (oospore formation) and post fruiting regeneration.

Objective 3 is to compare the spectral quality and fluence rates of light with filters and a quantum sensor (Licor) at the lower boundaries of *Nitella* meadows in New York State.

Objective 4 will attempt to determine the significance of the *Nitella* meadows as a phosphate sink The first experiments are designed to discriminate between the water and the sediments as the source of phosphate for the growing shoots.

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Title:
Insoluble Polymeric Contact Disinfectants for Small Water Treatment Systems

Principal Investigator:
Gilbert E. Janauer

Institution:
State University of
New York-
Binghamton

Project Period:
2 years

Budget:
(2 years) $138,144

EPA Project Number:
R 808265

The primary objective of the proposed research is to explore further a new approach to water disinfection, conceived in the laboratory of the Principal Investigator, to the point of making it feasible as an effective, maintenance-free method applicable to diverse small water treatment systems in the United States. The entire treatment will consist essentially of a single passage of natural (surface or well) water through a column or cartridge at the point-of-use, although other configurations (e.g., contact in storage tanks) appear promising also. The materials to be used—at the focal point of this research—are Insoluble Polymeric Contact Disinfectants (IPCD). They resemble in their general properties conventional ion exchange resins, but carry very high concentrations of animicrobial quaternary ammonium groups such as dodecyl ("Resin 12"), tetradecyl ("Resin 14"), or other suitable functions permanently attached to a stable, crosslinked polymer backbone. Efficacy against gram-positive and gram-negative bacteria, against several viruses, fungi, and algae has already been established in the laboratories of the Principal Investigator, but a number of intermediate research objectives must be attained in order to develop a practical method.

Standard procedures are to be finalized which permit quantitative comparison among different IPCD regarding their potency to disinfect, under real-life conditions, when compared with classical, soluble water disinfectants (e.g., iodine). The "best" IPCD with respect to contact time (flow-rate), disinfectant capacity (i.e. , total microorganisms disinfected before viable cells appear in effluent) as a function of pH, BOD, volume treated, resistance to water hardness and fouling by common trace organics, and ease of regeneration need to be determined before systemic testing toward a pre-prototype can be begun. The quality of treated water emerging from IPCD beds is to be assured by analysis for cell lysis products and any other potential contaminants at below "safe" levels. Physical-chemical and structural characterization of currently available IPCD resins is to be completed, and—based on these results, plus certain fundamental and practical considerations—new IPCDs with, possibly, even better performance are to be synthesized. The mechanism of disinfection action should be studied, because even a partial understanding will aid in the design of superior IPCDs (e.g., by exploiting potential synergistic effects). The anticipated disinfectant action against protozoa is to [illegible] tested also. Finally, a simple device—such as being used in household water treatment units—is to be adapted for actual field tests.

The microbiological methods used are standard or straightforward except for the comparative-efficacy protocol to be developed which may be based on a "rate-of-kill" approach similar to that outlined by Isquith et al. Characterization of IPCD materials will be carried out by classical methods (capacity, elemental analysis, scanning electron microscopy, and by IR and NMR spectroscopy. Treated water will be analyzed by GC-MS and, possibly, HPLC following preconcentration. Synthetic methods will be essentially those employed in conventional ion exchange resin synthesis except for surfactants used, and control of temperature and solvent which appear to be critical.

Title:
Effect of Pollutant Interaction on Environmental Transfer and Distribution

Principal Investigator:
Sujit Banerjee

Institution:
Syracuse Research Corporation

Project Period:
2 years

Budget:
(2 years) $120,383

EPA Project Number:
R 808613

The objective of this research is to measure the solubilities and bioconcentration factors of various chlorobenzene mixtures in fish. It is anticipated that the solubilities of components in a mixture will be progressively diminished as the complexity of the mixture increases. Variations will be quantified and an attempt will be made to rationalize them with the assistance of various theoretical models. Finally, the bioconcentration in fish of several mixtures of different concentrations will be measured in order to determine whether a similar depression occurs in mixtures.

A goal of this project is to demonstrate that environmental assessment based on individual compounds is erroneous, and that mixtures of chemicals must be considered as they occur in the environment. It is anticipated that, for the most part, the research will demonstrate that mixtures of chemicals are less hazardous than would be expected from consideration of their components. In this event, the research will be a step towards reducing the cost of regulation.

Title:

Investigations of the Effects of 20 Priority Pollutants on the Bacterial Disease Resistance of Rainbow Trout

Principal Investigator:
Richard H. Sugatt

Institution:
Syracuse Research Corporation

Project Period:
1 year

Budget:
(1 year) $38,940

EPA Project Number:
R 809004

Evidence has accumulated that acute or chronic sublethal exposure to water pollutants may lower the resistance of fish to infectious disease. In addition, there is some evidence that disease resistance may be as sensitive as chronic survival, growth or reproduction as an indicator of sublethal toxicity for some toxicants.

The main objective of the research program is to determine the effects of acute sublethal exposure to 20 priority pollutants (metal compounds, pesticides, synthetic organic chemicals, energy-related hydrocarbons) on the disease resistance of rainbow trout (*Salmo gairdneri*) against a facultative bacterial fish pathogen (*Aeromonas hydrophila*). Other objectives of the research program are to determine the static 96-hour LC_{50} of these priority pollutants, to develop a reproducible test system which can be used in conjunction with routine acute toxicity tests to measure the effects of acute sublethal pollutant exposure on the disease resistance of fish, and to assess the sensitivity of this test system as an indicator of sublethal toxicity by determining whether disease resistance is affected at exposure concentrations lower than the LC_{50} for a wide variety of pollutants.

The general methodological approach is to expose groups of trout to a range of test chemical concentrations (designed to include the 96-hour LC_{50}) under static exposure conditions, and then to compare the disease resistance of surviving fish at each toxicant concentration with that of unexposed control fish by exposing all groups to bacteria. The 96-hour LC_{50} will be determined according to standard methods recommended by EPA and ASTM, using duplicate groups of 20 trout at each toxicant concentration and control. At the end of the four-day exposure period, all tanks will receive clean flowing water to remove the toxicant. The water will be shut off and one of the duplicate groups at each toxicant concentration and control will be challenged by adding bacteria directly to the water. After 30 minutes, the water will be turned back on. The unchallenged duplicates will serve as additional controls to account for delayed toxicant-induced mortality. Disease mortality and toxicant-induced delayed mortality will be determined for an additional week, during which time all groups of fish will be fed and maintained under flow-through conditions.

This research will serve to assess the potential impact of 20 priority pollutants on fish disease resistance and will establish 96-hour LC_{50} values for the test compounds, many of which have not been determined previously with rainbow trout. In addition, this research may provide a standardized and ecologically realistic sublethal effects testing procedure which can be used to enhance the toxicological information obtained from routine acute toxicity tests at little additional cost.

Title:
Effect of Environmental Variables on PCB Biodegradation

Principal Investigators:
G. S. Sayler,
R. Pagni,
R. Bryant

Institution:
University of
Tennessee

Project Period:
3 years

Budget:
(2 years) $140,000

EPA Project Number:
R 808457

This investigation seeks to determine the interrelationships among biotic and abiotic factors contributing to the uptake and degradation of polychlorinated biphenyls (PCBs) by natural bacterial populations. The goal of this investigation is to provide needed information to develop a predictive model for assessing the environmental fate of biorefractile contaminants.

The specific objectives of this research are:

(1) To determine the kinetics of PCB photodecomposition in natural surface water.
(2) To determine the rate and mechanisms of PCB accumulation and biotransformation by natural microbial populations.
(3) To evaluate synergistic effects of photodecomposition and biotransformation on the environmental fate of PCB.
(4) To evaluate the role of dissolved organic matter, dissolved oxygen, particle surfaces, and temperature on modulating the environmental fate of PCB and the toxicological significance of PCB transformation products.

The methods used in this investigation include cultivation of genobiotic and natural bacterial populations in the presence and absence of photosensitized, pure or mixed, ^{14}C-labeled PCB cogeners. Following separation of the bacterial population from the aqueous media, ^{14}C-PCB accumulated by bacteria is quantitated by liquid scintillation spectrometry. The aqueous media are extracted with organic solvents and the extracts are analyzed by co-chromatographic, gas-liquid and high performance liquid chromatography techniques, and mass spectrometry to isolate, quantitate and identify the parent substrate and transformation products. Transformation products are evaluated for toxicological significance using bacterial and mammalian mutagenesis assays. The role of environmental variables in modulating the fate of PCB is assessed by manipulating experimental variables in laboratory cultivation and microcosm experimentation.

Title:

The Effect of Exposure to Texas Gulf Coast Air Pollution in Asthmatics

Principal Investigator:
Alfonso H. Holguin
Institution:
University of Texas, Health Science Center at Houston

Project Period:
17 months

Budget:
(1 year) $901,736

EPA Project Number:
R 808738

Several epidemiologic studies have demonstrated that increased concentrations of several types of ambient pollutants, notably ozone, have been associated with an increased number of asthmatic attacks, or with an increase in the severity of attacks. Since the Texas Gulf Coast area not only experiences high but variable levels of many types of pollution, an unusual opportunity is provided to compare changes in asthma status with changes in the pollution level and composition of the ambient air. This study has been designed to permit investigators to detect associations between changes in the biomedical status of asthmatic subjects with changes in environmental factors that include pollutants and meteorologic variables.

Daily symptomatology, lung function, and medication use were measured in a selected group of asthmatics in Houston during the summer of 1981. Concurrently, levels of air pollutants to which the asthmatics were exposed were recorded.

The data collection phase generated two major data sets: (1) biomedical data and (2) air monitoring data.

(1) Biomedical Data. Fifty-two physician-diagnosed asthmatics, aged seven years and older, participated in a six-month study. One-half resided in one of the study areas, the other one-half in the other study areas. The period of observation chosen was that of highest expected ozone concentrations, May through October.

Each participant maintained two log forms each day, covering a 12-hour period. Information gathered on the logs included medication use, subjective symptomatology, activities, and any episodes of respiratory distress requiring instructions and/or assistance from a physician. The subjects use Mini-Wright Peak Flow Meters to measure and record the results of three successive trials done when completing their log forms. Peak flow meters were calibrated weekly, and the participants questioned to determine any changes in residence or workplace or physiological state which might affect their asthma.

(2) Air Monitoring Data. A fully-instrumented fixed-site air monitoring station was located at the center of each study area. The fixed sites sequential integrated sampling was performed for total suspended particulates, inhalable particulates, aldehydes, and aeroallergens. In addition, particulate filters were routinely analyzed for sulfate, nitrate, and trace elements. Special samplings were performed on a regular basis for the determination of aerosol acid and carbon content.

A mobile van, equipped with the same monitoring instrumentation as the fixed site monitoring station, was employed to measure indoor and outdoor pollutant levels in a representative sample of the asthmatics' homes. Week-long integrated estimates of personal exposure to nitrogen dioxide and formaldehyde were obtained by having the subjects wear small passive diffusion badges. Exposure to respirable particulate and ozone were periodically measured by technicians with portable monitors accompanying the study participants through their daily routines.

Associations between health effects observed, the biomedical data and levels of selected pollutants are currently being investigated; first using single pollutants, and then employing pollutants in combination.

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Title:
Cytogenic Effects of Chemical Mutagens on Spermatogenesis

Principal Investigator:
T. C. Hsu

Institution:
University of
Texas Systems
Cancer Center

Project Period:
3 years

Budget:
(2 years) $414,744

EPA Project Number:
R 808582

There are two major types of environmental mutagens: (1) those which cause DNA damage and mutations (mutagens) and chromosome breakage (clastogens), and (2) those which interfere with cell division mechanisms (mitotic poisons). A variety of test protocols have been developed for the former, from microorganisms to experimental animals *in vivo*. However, test systems for the latter have been deficient.

Regular cell division in eukaryotes distribute the genetic material (chromosomes) equally from a mother cell to the daughter cells. Many cellular organelles are involved, including the spindle (microtubules), the centrioles, the kinetochores, and the cytokinetic mechanisms. Abnormalities occurring in any of these structures may lead to polyploidization and aneuploidization. In view of the occurrence of aneuploid individuals in the human population and the preponderence of aneuploidy in cancer cells, this class of genotoxic substances should be vigorously tested. This project proposes to develop cytogenetic test systems using cells of mammalian and nonmammalian materials.

Known and suspected mitotic poisons will be tested in the following materials:

(1) Laboratory mice. The mice will receive intraperitoneal injections of the test compound, and be sacrificed various days postinjection. Bone marrow and testes will be procured for cytogenetic preparations to study divisional anomalies and aneuploidy, if any. The testicular material is more important because the end product of gametogenesis represent the only link between parents and offspring. Major target cells for scrutiny include metaphase I, metaphase II, and spermatid development.

(2) Other test systems using insects (grasshopper embryos and cockroach testes) are being developed because these systems show promise.

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Title:
Determination of Nekton Community Structure in Shallow Marsh Habitats

Principal Investigator:
Michael Weinstein

Institution:
Virginia Commonwealth University

Project Period:
1 year

Budget:
(1 year) $47,393

EPA Project Number:
R 808707

This research attempts to assess the role of tidal creeks in the production of estuarine fish and shellfish important to man—within the framework of community structure and function. General objectives of the program include:

(1) Determining how food resources are partitioned within the creeks.
(2) Determining how space is utilized in these habitats.
(3) Determining periods of residency and population turnover rates for individual species.
(4) Estimating the relative contributions to secondary productivity of brackish and high salinity tidal creeks.
(5) Assessing the relative roles, as primary nurseries, of marsh creeks and seagrass meadows.

The early life stages of fish and shellfish will be collected from two well-defined tidal creeks in different salinity regimes. These creeks will be small enough to allow them to be collected relatively intensively along their length. A multiple gear approach is proposed for this study with a uniform effort distributed in each creek to allow direct comparisons between areas. The primary sampling methods will consist of replicate trawls of a semi-balloon otter trawl and replicate tosses of a modified Wegener ring (in the narrow, upper creeks). For each monthly (or seasonal semimonthly) collection pertinent water quality data will be determined.

Gut contents of randomly selected subsamples of individual species will be identified in the laboratory and several methods will be used to record these data including enumeration, biovolume and dry weight measurements and several indices or combinations thereof will be used to express the results. The composition of all collections will be recorded (species and abundance) and the data compared between creeks, among sites within creeks and among seasons in a factorial design. These data are also amenable to several community analysis procedures.

Population dynamics of selected species also will be examined via an intensive marking program utilizing "sandblasting" with fluorescent pigments. Production estimates for these same species will be derived from both the marking study and the overall sampling program.

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Title:
Cellular Effects of Radiofrequency Radiation

Principal Investigator:
S. F. Cleary

Institution:
Medical College
of Virginia

Project Period:
3 years

Budget:
(3 years) $192,433

EPA Project Number:
R 808617

The objective of this investigation is to determine the effects of radiofrequency radiation (RF) in the frequency range of 100-200 MHz on specific cell functions *in vitro* and to ascertain the extent to which RF-induced functional alterations are reversible (i.e., repairable). Studies are being conducted on mammalian blood cells (erythrocytes, polymorphonuclear leukocytes, and lymphocytes), transformed neural cells cultures (glioma and neuroblastoma) and normal (untransformed) diploid human embryo fibroblasts. The specific cell functions being investigated include: cell morphology, surface characteristics, viability, plasma membrane cation permeability (K^+, Na^+, Ca^{++}), proliferative response, phagocytosis and transmembrane potential. A long-range objective of this investigation is to provide insight regarding the mechanisms of interaction of RF radiation with cells that results in such functional and/or structural alterations.

Cells will be exposed to RF radiation in coaxial transmission line cell culture sample holder and heat exchanger. Exposures will be at constant, controlled temperatures at field strengths of up to 30V/cm. Exposure effects of mammalian blood cells in suspension and cultured neural cells (rat glioma C6 and mouse neuroblastoma NB2A) and fibroblasts will be determined as a function of RF frequency, intensity, amplitude-modulation, exposure duration, temperature, dosage mode, and cell density.

Title:

Investigation of the Transport and Diffusion of Pollutants in Complex Terrain Using a Numerical Mesoscale Model

Principal Investigator:
R. Pielke

Institution:
University of Virginia

Project Period:
2 years

Budget:
(2 years) $70,607

EPA Project Number:
R 807813

The overall objective of this research project is to utilize a primitive equation mesoscale model to simulate the atmospheric flow patterns induced by complex terrain and examine the impact these flow regimes have on the transport and diffusion of air pollutants. A more specific objective is to apply the mesoscale model research to EPA's planned field program for terrain impaction.

As part of the overall objective, a detailed evaluation of the model-predicted thermal circulations will be made against available experimental data. Model velocities, scales and timing will be compared to experimental values. Recently available detailed observations using tethered balloons may be quite useful in evaluating the vertical structure of the flow.

The Lagrangian conditioned particle scheme for point sources of pollution will be tested further. Additional experiments will be made to examine the accuracy of current methods for the extraction of the turbulent parameters, $(\sigma)_w$, $(\sigma)_{u,v}$ from the exchange coefficients produced by the mesoscale model. New information on turbulent scales and boundary layer structure in complex terrain will be useful for model validation and model parameterization of length scales.

Using the Lagrangian particle model, two and three-dimensional mesoscale model runs will be made for idealized terrain obstacles. Particle puffs from various heights will be released so as to encounter the terrain obstacle. The role of wind shear and the influence of the turbulent boundary layer developing over the terrain obstacle on particle dispersion will be investigated.

Some resources will continue to be given to improvement of the mesoscale model specifically in the area of the parameterization of turbulent exchange processes. Numerical improvements in the mesoscale model and efficiency of the computational processes will also be given some attention. Computational costs can undoubtedly be reduced by some restructuring of the model code. Although the areas to be given attention will depend upon problems encountered and by advancements made by other members of the research group at the University of Virginia, some possible candidates for improvement are: radiative boundary conditions, meshed grids (as opposed to current model stretched grids), simplifications to the surface energy budget, and modified spline advection techniques. Depending on the progress of the project, an investigation of the scales to which the hydrostatic model is valid under stable conditions may be carried out.

Title:
An Atmospheric Tracer Investigation of Flow and Dispersion Near an Isolated Building

Principal Investigators:
Brian Lamb,
Elmer Robinson

Institution:
Washington State University

Project Period:
1 year

Budget:
(1 year) $52,357

EPA Project Number:
R 808446

The dispersion of pollutants released from buildings and the turbulence patterns of the atmosphere near buildings will be investigated by performing a series of atmospheric tracer experiments around an isolated industrial building. Continuous meteorological measurements in the prevailing upwind and downwind sectors will be conducted throughout the study period. During typical tracer tests, one tracer ($CBrF_3$) will be released upwind of the building in the undisturbed flow region while a second tracer (SF_6) is released from the roof of the building into the disturbed flow region downwind of the building. Comparison of horizontal and vertical dispersion rates for the undisturbed plume and the disturbed plume will allow the effects of the building-generated turbulence to be separated from the effects of natural atmospheric turbulence upon plume dispersion. The tracer data will be used to verify results from similar scale-model wind tunnel studies and to test the validity of current industrial source dispersion models. This field investigation will provide further data for studying relationships among building dimensions, recirculation cavity dimensions, atmospheric turbulence levels, and pollutant concentration fields near the building.

Title:
Stability Classifications for Air Pollution Dispersion Models

Principal Investigator:
Elmer Robinson

Institution:
Washington State University

Project Period:
2 years

Budget:
(2 years) $108,069

EPA Project Number:
R 808702

The objective of this research study is the development of a revised procedure to use in order to estimate the stability categories that are necessary inputs to air pollutant transport and diffusion models. These revised procedures should make use of standard National Weather Service observational data so that they can be used as easily as currently used methods developed by Pasquill in 1961 and Turner in 1964 and which are now incorporated in the stability climatology computer program STAR.

The problem with the STAR-type data, as identified by a number of investigators, is inappropriate stability identification under conditions of strong solar radiation and wind. The present research program hopes to address this situation by considering additional data inputs such as a temperature function.

The research plan includes verification programs in which any new stability identification procedure could be compared in terms of performance with the STAR system and other currently available techniques.

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Title:
Toxicant Removal in Submerged Media Anaerobic Reactors Processing Industrial Waste

Principal Investigators:
John F. Ferguson,
Mark M. Benjamin

Institution:
University of
Washington

Project Period:
2 years

Budget:
(2 years) $153,963

EPA Project Number:
R 808675

The use of the high-rate anaerobic treatment of waste streams from the pulp and paper industry will be demonstrated. Kraft digester condensates, thermomechanical pulping effluents, and bleach plant effluents will be treated in pilot-scale reactors.

These wastes are particularly suited to anaerobic waste treatment due to their high concentrations of anaerobically biodegradable materials. The relationship between performance, degradable and non-degradable organics, detention time, and process loading will be assessed in a series of steady state tests. However, they also contain a number of compounds that may be toxic to methanogenic cultures. These compounds may inhibit methanogenesis initially, but acclimation is expected. Their removal by processes leading to destruction is also expected.

The study will assess the fate of a number of potentially toxic, substituted aromatic compounds in anaerobic processes. The toxicity, biodegradability, volatilization and sorption of these compounds will be determined in pilot-scale reactors. Carbon-14 labeling and gas chromatography will be used to trace the compounds of study through the anaerobic digestion process. These tests will also be used to relate structure and substitution to fate in the anaerobic process. GC/MSA analyses will also be used to survey these waste streams and assess treatment performance with respect to removal of certain trace constituents.

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Title:
Systemic and Cardiovascular Effects of Chronic Barium Ingestion

Principal Investigator:
H. Mitchell Perry, Jr

Institution:
Washington University School of Medicine

Project Period:
2 years

Budget:
(2 years) $160,505

EPA Project Number:
R 809175

Measurable barium is present in more than 99% of water supplies in the United States. The "maximum permissible concentration" is 1 milligram per liter. Areas exist in Illinois, Kentucky, Pennsylvania, and New Mexico where the concentration is ten times this high. A recent retrospective epidemiological study suggests that high barium concentration (1.1 to 10.0 ppm barium) in the drinking water of certain Illinois communities may be related to the significantly higher cardiovascular mortality rate in these communities from 1971 through 1975.

The barium ion causes intense stimulation of muscle, both smooth and striated. Cardiovascular effects include arrhythmia, spasm of arteriolar musculature, and arterial hypertension. A unique feature of acute barium toxicity is the marked increase in intracellular potassium and dramatic hypkalemia.

Prior *in vitro* work has demonstrated that spirally-cut rabbit aorta contracts in response to varying concentrations of barium and of several other vasoactive metals. *In vivo* studies have observed a marked pressor effect in rats from intra-arterially injected barium, with lesser effects from other vasoactive trace metals.

The present proposal is designed to examine physiologic, metabolic, and morphologic changes resulting from chronic exposure to ingested barium in a search for evidence of overt or latent health effects associated with barium.

Weanling Long-Evans rats will be maintained in "low contamination" animal facilities. They will be individually weighed monthly for four months and then bimonthly. At 1, 2, 4, 8, 12 and 16 months blood pressure will be measured. At 1, 2, and 16 months, one-third of the rats will be sacrificed. At sacrifice, His bundle electrograms will be obtained, and myocardial contractility measured. In addition, hematocrit and plasma electrolytes, organ weight and histology, and tissue concentrations of Ba, Ca, Mg, Na, K (and Cd or Pb when appropriate) will be obtained; finally plasma epinephrine and norepinephrine will be estimated in one-half of the animals.

Title:
Removal of SO_2 with Ferric Ion as a Catalyst

Principal Investigator:
Eung Ha Cho

Institution:
West Virginia University

Project Period:
2 years

Budget:
(2 years) $57,032

EPA Project Number:
R 809597

The objective of this research is to investigate the details of the mechanism and the kinetics of the SO_2 removal reaction using ferric ion as a catalyst. The effect of O_2 on the catalytic action will be thoroughly studied. Also, the kinetic data of the reaction will be analyzed according to hydrodynamical theory. The results of the experimental runs will be analyzed to yield information as to the proposed process effectiveness. The practical application of this system will also be assessed.

The research effort on the SO_2 removal will be divided into two major sections (1) the catalysis study, and (2) kinetic study. The experimental apparatus for the catalysis study will consist of a one-liter capacity reactor, temperature controller, and reflux condenser. Experimental variables to be applied for this system are concentration of ferric ion, solution pH, concentrations of SO_2 and O_2 in the gas stream, temperature, flow rate, and stirring speed. The experimental data will be analyzed to yield information on the catalytic mechanism of ferric ion in the reaction.

The kinetic studies will be carried out in a single drop reaction cell. The apparatus will consist of a reaction cell, an orifice for bubble formation at the bottom of the cell, SO_2 and O_2 gas tanks and its channel connected with the orifice and an SO_2 analyzer connected with the top of the reaction cell. The experiments will be designed so that the kinetics of the removal could be investigated with the rising drop. Experimental variables to be applied for this system are concentrations of SO_2 and O_2 in the gas phase, concentration of ferric ion, bubble size, contact time, and solution pH. The results will be analyzed according to the mass transfer models based on the hydrodynamics of the system.

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Title:

The Pathology and Microbiological and Integrated Control of Eurasian Watermilfoil in Relation to Water Quality

Principal Investigator:
John H. Andrews

Institution:
University of Wisconsin-Madison

Project Period:
3 years

Budget:
(2 years) $136,985

EPA Project Number:
R 807127

Eurasian watermilfoil (*Myriophylum spicatum* L.), a submersed aquatic angiosperm, is an aggressive invader that is currently causing severe problems in many regions of the United States due to its high reproductive capacity and extraordinary competitive ability. The main problems that arise from disruption of the aquatic plant community are impeded water flow, interference with navigation or recreation, clogging of water intake structures, and deleterious effects of decaying plant parts on water quality. Conventional control methods, including mechanical and chemical approaches, have been expensive and only partly successful. To surmount these difficulties, microbiological and integrated control of the weed will be attempted by manipulation of normally occurring microorganisms pathogenic to it in coordination with other methods.

Objectives

(1) To study the impact of water quality factors on the pathology of milfoil in Wisconsin.
(2) To determine microbiological, mechanical and chemical components for a coordinated program.
(3) To determine the impact of integrated control on water quality.

Milfoil grown in lighted aquaria will be subjected to pathogens and to various regimens of water chemistry and quality including sediment type, relative hardness, nutrient status, turbidity, and temperature. Effect of these variables on pathogenesis will be assessed by symptom development; effect on the host will be determined by measuring biomass as dry weight. Juvenile milfoil in aquaria will be subjected to mechanical harvesting, chemical or biological control with pathogens, or to all possible combinations of these treatments. Efficacy will be assessed as above. Promising additive or synergistic controls will be tested further in experimental ponds or in large plastic enclosures in the lake. Water quality parameters used to measure impact of control strategies will include survival and host specificity of the pathogen, seasonal periodicity of phytoplankton, nutrient levels as represented by phosphate, turbidity, pH, and biochemical oxygen demand. Pilot studies will be conducted *in vitro*. Later trials will be in experimental ponds or within plastic enclosures in the lake.

Title:
Evaluation of Sewage Treatment Plant Performance

Principal Investigator:
P. M. Berthouex,
W. G. Hunter

Institution:
University of Wisconsin-Madison

Project Period:
2 years

Budget:
(2 years) $157,930

EPA Project Number:
R 809162

The objective of this research is to define the capabilities of treatment plants to perform consistently. Existing records of effluent quality cover a combination of intervals when plants function as well as they should and other intervals when they perform poorly because of operator error, equipment malfunction, or other disturbances. A typical study of effluent quality condenses a plant's effluent quality record into a frequency distribution and uses that distribution to define satisfactory performance. A study is needed that segregates and carefully examines periods of poor treatment in an attempt to find assignable causes for such unsatisfactory behavior. Then the dynamics of the process during normal operation could be statistically characterized. Records from a number of treatment plants will be analyzed, including a number of plants that have been studied by others.

Simply making the frequency distribution analysis of effluent quality is not sufficient to achieve the objectives. Neither is the application of standard quality control theory because some of the fundamental important assumptions underlying the usual quality control methods are violated by treatment plant operating data. A combination of quality control methods and time series techniques is needed to identify portions of records that are thought to be in-control or out-of-control. These two separate portions then will be studied carefully. Neither statistical method alone is adequate for this purpose.

This research will (1) improve understanding performance variation, (2) provide a baseline of performance against which to compare proposed innovative designs, (3) improve operational reliability by more precisely defining performance expectations, and (4) develop methodology to permit others to carry out similar analyses on other sets of data.

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Title:
Trihalomethanes in Drinking Water and Colon Cancer

Principal Investigator:
Marty Kanarek

Institution:
University of Wisconsin-Madison

Project Period:
3 years

Budget:
(2 years) $224,954

EPA Project Number:
R 808685

In response to preliminary epidemiologic and laboratory studies which suggest an association between exposure to chlorinated drinking water and cancers of the urinary and digestive tract, a case-control study of environmental factors and cancers of the colon and rectum is being conducted in Wisconsin. The primary objective is to determine the magnitude and significance of cancer risk associated with estimated lifetime ingestion of chloroform, total trihalomethane, and total chlorinated volatiles. In addition, the risk associated with other water contaminants, dietary habits, occupational and other life style factors will be explored.

The data base for comparing case exposures to control exposures will be assembled from responses on a mail-back questionnaire given to a statewide sample of 1600 incident cases of colon and rectum cancer and 3200 controls (one set of non-digestive, non-urinary cancer controls, and one set of general population controls). The data base for comparing case exposures to control exposures will be assembled from merging (1) residential histories and information from a mail-back questionnaire, on water drinking and bathing habits, (2) estimates of drinking water exposures which prevailed at the past geographic locations where each subject resided. The estimates for each water source will be derived by a statistical modeling procedure in which quantitative, seasonal measurements of chlorinated by-products in the finished water of 100 waterworks will be regressed on water parameters for which historic data are available (chlorine dose, pH, source type, etc.). It will then be possible to use the historic data available from waterworks records (appropriately weighted) to estimate the drinking water exposures for all sources at any point in time.

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Title:
Human Cell Activation of Polycyclic Aromatic Hydrocarbons

Principal Investigator:
Leila Diamond

Institution:
Wistar Institute

Project Period:
2 years

Budget:
(2 years) $342,150

EPA Project Number:
R 809202

The objective of this project is to determine the validity of using established human hepatoma cell lines in culture as activators for polycyclic aromatic hydrocarbons (PAHs) in short-term genotoxic assays that require an exogenous activation system. The project is based on our preliminary evidence that one such cell line that we have isolated, HepG2, can activate two PAH carcinogens, benzo(a)pyrine (BP) and 7,12-dimethylbenza(a)anthracene (DMBA), to mutagenic metabolites in a cell-mediated assay with V79 Chinese hamster cells as target cells. The protocol also will provide information about the potential use of SENCAR mice, which have been specifically bred for susceptibility to PAH carcinogenesis, in *in vivo* screening assays for PAH carcinogens.

Using the HepG2 cell line as an activation system in cell-mediated assays and selection of 6-thioguanine-resistant V79 target cells as the marker, a series of PAH of known genotoxic (carcinogenic) and non-genotoxic activity will be tested first for mutagenic activity. Then a series of PAH derivatives with fluoro or methyl substitutions at various positions on the molecule and unknown mutagenic or carcinogenic activity will be tested in the *in vitro* assay. This latter group of compounds also will be tested for skin tumor initiating activity in SENCAR mice using an initiation/promotion protocol. To determine if HepG2 cells (or other human hepatoma cell lines that will be tested) are *more* appropriate as activators than rodent cells or tissue preparations for predicting genotoxic or carcinogenic potential of individual PAH for humans, the metabolism of BP and DMBA by HepG2 cells and normal human liver fractions will be compared to metabolism by rat hepatocytes and rat liver fractions. Hydrocarbon-DNA binding in V79 target cells mediated by the various cells and tissues also will be compared.

These studies will help to validate the HepG2 cell-mediated mutation assay for PAH, increase our knowledge of species extrapolations for PAH activation and of the structural requirements of hydrocarbon molecules for mutagenic and carcinogenic activity and provide additional information on the use of SENCAR mice in "short-term" *in vivo* assays for PAH carcinogens.

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Title:
Catalytic Air Oxidation of Industrial Effluent

Principal Investigator:
Gerald S. Vigee

Institution:
University of Alabama in Birmingham

Project Period:
2 years

Budget:
(2 years) $124,087

EPA Project Number:
809726

The treatment of industrial waste water containing pollutants such as phenols is expensive and energy consuming. Treatment methods presently available utilize carbon adsorption, chemical means (condensation of phenols with aldehydes) and biological agents which are found to metabolize (oxidize) phenol substrates are phenoloxidases, metalloenzymes which possess binuclear copper(11) ions at the active site. The enzymes use aerobic oxygen as oxidants and the reaction is significantly enhanced by sunlight.

The object of this project is to prepare and test polymeric copper catalysts which exhibit oxidase activity and can be used to treat phenols in waste water. The complexes are expected to mimic the oxidase behavior of the metalloenzymes found in microbial sludges presently used in the treatment of phenol containing waste water but will lack the inherent shortcomings of the living microbial agents.

Polymeric copper complexes, similar to the simple copper complexes exhibiting oxidase activity which were previously prepared in the author's laboratory, will be prepared and tested for oxidase activity against phenols in water. The complexes will be anchored to prevent their loss during oxidase treatment. They will be tested for oxidase activity under varying conditions of pH, light and dark and their ability to precipitate out polymeric phenols by flocculation will be investigated.

It is expected that the simple metalloenzyme-like complexes will effectively remove or neutralize as much soluble catechols or phenols as the presently used biological agents without the disadvantages found in the use of biological agents. Only laboratory testing is planned in this project. Scale up to pilot plant size testing should present little difficulty.

Title:

An Empirical Model for Predicting Trihalomethane Formation in Synthetic and Natural Waters

Principal Investigator:
Gary Amy

Institution:
University of Arizona

Project Period:
2 years

Budget:
(2 years) $112,942

EPA Project Number:
R 809935

This research project represents a systematic study to describe the kinetics and extent of trihalomethane formation during the chlorination of synthetic and natural waters containing humic substances. The objectives of the proposed study are (1) to develop an empirical model which can predict trihalomethane formation as a function of time, based on a series of initial water quality characteristics and reaction conditions, (2) to quantitatively assess the effects of precursor type, purcursor concentration, chlorine dose, temperature, pH, and bromide concentration on the kinetics and ultimate yield of trihalomethanes, and (3) to ascertain the effects of several candidate treatment processes on THM reaction kinetics and yield.

The formation of trihalomethanes during chlorination is being investigated for a broad spectrum of both synthetic and natural water samples containing humic substances. The effects of various degrees of water treatment on the kinetics and yield of trihalomethanes is being evaluated by considering several candidate treatment processes selected for precursor removal, including chemical coagulation, activated carbon adsorption, and ozonation. Trihalomethane formation is being studied in both untreated and treated waters by conducting batch-mode experiments in which a wide range of experimental conditions (i.e., temperature, pH, etc.) are evaluated.

Multiple linear and nonlinear regression are being employed to statistically analyze groups and subgroups of data in order to quantitatively represent the trihalomethane formation reaction. Data corresponding to each unique water are being evaluated individually to develop a series of empirical submodels applicable to each of the waters. Eventually the entire data base will be used in attempting to formulate an overall model. Finally, the overall model will be modified to account for changes in precursor characteristics induced by treatment, thus enabling prediction of THM formation in various waters following treatment.

Title:
Detection of Rotavirus and Hepatitis A in Water

Principal Investigator:
Dr. Charles P. Gerba

Institution:
University of Arizona
Tucson, AZ

Project Period:
2 years

Budget:
(2 years) $116,945

EPA Project Number:
R 809855

Objectives

The primary objectives of this project are: (1) to improve methodology for the detection of rotaviruses and hepatitis A (HAV) virus in water; (2) to determine the ability of rotaviruses and HAV to survive domestic sewage and drinking water treatment; and (3) to determine the occurrence and concentration of rotaviruses and HAV in sewage, polluted recreational waters and reclaimed waters.

Expected Results of Research:

The previous absence of applicable methodology for virus detection in waters and the difficulty of epidemiologic studies have limited the amount of available information on waterborne viral disease. Because of the difficulty and cost of epidemiologic methods, it is felt that the presence of viruses in a water supply can be an indication of a disease threat. Waterborne outbreaks of viral disease have been documented for infectious hepatitis, enteroviruses, rotavirus, adenoviruses Norwalk-like agent, and other agents of non-bacterial gastroenteritis believed to be viral. An improved detection system for viruses would provide a means for early prediction of potential waterborne disease outbreaks.

Title:
Fuel Nitrogen Mechanisms in Laminar Opposed Jet Diffusion Flames

Principal Investigator:
Jost O. L. Wendt

Institution:
University of Arizona

Project Period:
2 years

Budget:
(2 years) $97,096

EPA Project Number:
R 809968

The principal objectives of this research are to identify and quantify the significant chemical reaction paths for the conversion of fuel nitrogen under strained, laminar, opposed jet, diffusion flame conditions. Thus, this work focuses on the chemical events occurring in the first few milleseconds of reaction history under non-premixed, diffusion conditions. An understanding of this short time-constant behavior is required to predict the effects of fuel nitrogen type, fuel type and NO reprocessing by flames, and should aid in the development of inexpensive NO_x control strategies through combustion modification.

The laminar opposed jet combustion configuration has a number of unique attributes: first, it can be modeled in detail with finite rate elementary reactions; second, it is a clean experiment, both analytically and experimentally, because influences of boundary surfaces are minimized and ideal conditions can be approached; third, it allows the study of kinetics in extremely rich pyrolysis environments; fourth, the strained flame is representative of flamelet environments embedded within large structure turbulent diffusion flames. Thus, the kinetic routes uncovered in this work have some bearing on those important in practical turbulent diffusion flames.

The project consists of two phases. In the first phase, focus is on reaction paths important in the processing of fuel nitrogen in CO/H_2 flames without hydrocarbons. These studies are, therefore, with a fuel whose reaction mechanism and free radical production are understood and this will allow simplifications in interpreting fuel nitrogen pyrolysis and oxidation mechanisms. In phase 2, hydrocarbons will be added to the fuel, but in dilute amounts in order to prevent the formation of soot. Extractive sampling and optical techniques will be used to measure stable and some transient species. In all cases the objective is clearly to identify fuel nitrogen reaction paths with as few obscuring factors as possible.

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Title:
A Multi-Species Comparison of the Neurobehavioral Toxicology of Alkyltin

Principal Investigator:
Donald E. McMillan

Institution:
University of
Arkansas for
Medical Sciences

Project Period:
3 years

Budget:
(2 years) $287,022

EPA Project Number:
R 809452

Alkyltin compounds are used widely in industry and represent an important class of neurotoxins. Both triethyltin (TET) and trimethyltin (TMT) produce lesions in the hippocampus. TET has been shown to produce a disruption of the myelin sheath with little neuronal involvement. Recently, however, TMT has been shown to produce a degeneration of the neuron with little involvement of the myeline sheath. The specificity of the lesions produced by TET and TMT provides a unique opportunity to correlate functional deficits with pathology. These tests will include spontaneous motor activity, and schedule-controlled (operant) behavior. The tests using the schedule-controlled behavior will include schedules which are sensitive to a wide variety of behaviorally active substances (i.e., multiple fixed-ratio, fixed-interval), and schedules which are useful for controlling behavior which may be related to hippocampal functions of learning and memory (i.e., single alteration, repeated acquisition, matching-to-sample). Neuropathology of TET and TMT administration will be studied in both the animals used in the behavioral studies and in animals treated identically but not used in behavioral species: mouse, rat and pigeon. Although the rat has been used extensively in previous studies with alkyltin, preliminary results in this laboratory has shown that the mouse and the pigeon are approximately 2-3 fold more sensitive. Thus, they may prove to be better animal models than the rat. In addition, the multi-species approach will add generality to our findings.

Title:

"The Spouted Bed: A Novel Approach to Burn Low Calorific Value Fuels in an Environmentally Acceptable Manner"

Principal Investigator:
Michael J. Murphy

Institution:
Battelle-Columbus Laboratories

Project Period:
1¼ years

Budget:
(1¼ years) $130,632

EPA Project Number:
R 809584

The overall objective of this program is to provide a quantitative basis for assessing the spouted bed combustor as a burner for low heating value fuels. Whereas conventional combustion systems use preheat to extend the range of flammability, the spouted bed incorporates internal heat recirculation to raise the adiabatic flame temperature. The spouted bed thus has the potential to burn low-grade fuels completely without an external source of energy or an external heat exchanger.

In order to investigate the possibilities of the spouted bed for burning such fuels, we have constructed a 150 mm diameter laboratory-scale spouted bed combustor. This lab-scale unit is equipped to burn solid, liquid, and gaseous fuels. A sample probe and sample line connected to a set of emissions measurement instruments allow the combustion products to be monitored.

Experiments will be conducted in which the heating value of the fuel is changed systematically in order to identify the lower limit of stability. In addition to measuring gaseous emissions, observations of the overall flame shape and structure will be made.

The results will include the fuel tolerance of the spouted bed, emissions data, fuel feeding procedures, and overall flame structure observations.

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Title:
Short-Term Tests for Chemical Nephrotoxicity

Principal Investigator:
Marjorie G. Horning

Institution:
Baylor College of Medicine

Project Period:
3 years

Budget:
(2 years) $410,275

EPA Project Number:
R 809590

The chief objective of the present study is to develop reliable short-term tests that can be used to test for nephrotoxicity of environmental compounds. Three groups of nephrotoxic compounds will be studied. Group I includes compounds known to damage the proximal convoluted tubule and to cause glucosuria. These compounds will be used to establish the validity of an experimental design based on failure of the glucose reabsorption mechanism. Compounds in Group II include chemicals that are nephrotoxic, but where there is not complete agreement with respect to the incidence of glucosuria and where lesions have been observed in the straight portion of the proximal tubule. Most of these compounds are halogenated and all are hepatotoxic as well. Group III includes substances that are regarded as nephrotoxic, but where the mechanisms of toxicity and site(s) of injury are not well defined. Inasmuch as some of the compounds (carbon tetrachloride and bromobenzene) are more nephrotoxic to mice than to rats, studies will be carried out both in rats and mice to determine the extent to which the glucose response is species, strain or sex dependent.

A second objective is to study the effect of nephrotoxic compounds on the excretion of endogenous organic acids. It is well known that many nephrotoxic agents alter anion transport. By comparing the 24-hr urinary acid profiles of control rats with profiles obtained from rats treated with nephrotoxic doses of the compounds under study, it may be possible to identify one or more changes that can serve as a marker for altered anion transport resulting from kidney damage. Characteristic changes in acid profiles have been observed following administration of hepatotoxic agents; it is not yet known if changes result from kidney toxicity.

A third objective is to investigate the ability of protective agents to prevent renal toxicity due to environmental chemicals. In preliminary studies, it was found that pretreatment with cobaltous chloride prevented the functional damage induced by large doses of salicylic acid and acetaminophen. These results indicate that glucosuria was caused by metabolism of the drugs to reactive intermediates by enzymes of the P450 type, and that these intermediates may have been formed in the kidney. The protective effect of N-acetyl-L-cysteine and the anti-oxidant BHA (butylated hydroxyanisole) on the incidence and severity of glucosuria induced by nephrotoxic compounds will be studied. The studies with protective agents should provide insight as to the nature of reactive intermediates and to the molecular pharmacology of nephrotoxicity caused by environmental compounds.

Title:
A Rapid, Sensitive and Inexpensive In Vivo *Carcinogenesis Test*

Principal Investigator:
Seymour Holtzman

Institution:
Brookhaven National Laboratory

Project Period:
2 years

Budget:
(2 years) $242,763

EPA Project Number:
R 810142

The aim of this proposal is to develop and validate a simple, rapid, sensitive and inexpensive *in vivo* screen for chemical carcinogens. The system is based upon the direct application of test materials in either solution, or suspension, or dry mixtures to specific sites in the breast tissue of female Sprague-Dawley rats, with a subsequent 90-day follow-up to determine mammary adenocarcinoma production. Each animal can be used for both dose response studies and as its own control because there are up to 12 sites (nipples) available for application. Thus, fewer animals and smaller quantities of test materials will be needed than are used in standard carcinogenesis tests. Promotion by increased dietary fat intake will be attempted to increase the sensitivity of the test. For chemicals which are negative in the test, or which are not considered to be mammary carcinogens, we will try activation with Arochlor-induced rat liver S-9 fractions. The screening test will be developed for both pure chemicals and complex mixtures. Materials that have already undergone a variety of *in vitro* and *in vivo* tests will be used for validation purposes. Aside from its intrinsic value for investigatory purposes by Federal regulatory agencies such as the EPA, and by Universities and other research organizations, the relative simplicity and low cost of this screen should make the test useful to state and local governments. The screen should also allow industries with limited finances and facilities to develop their products more rapidly and safely, while cutting costs to the consumer public.

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Title:

Intestinal Barrier to Particulate Matter in Young and Old Animals

Principal Investigator:
M. E. LeFevre, PhD

Institution:
Associated
Universities, Inc.,
Brookhaven National
Laboratory,
Upton, NY

Project Period:
3 years

Budget:
(2 years) $279,175

EPA Project Number:
R 810140

Objectives:

This project will attempt to quantitate penetration of the intestinal mucosal barrier by non-digestible particulate matter in weanling, young adult, and old mice. Test particles, 1.5-μm fluorescent and 5.7-μm nonfluorescent latex will be administered chronically by gavage. ^{7}Be-labeled carbon (a new test particle produced by bombardment of carbon black with high-energy protons) will be given by gavage in single-dose experiments. Morphology of the tissues involved will be studied by light and electron microscopy with emphasis on the use of Peyer's patches, the region of the small intestine known to be the major site of particle penetration.

Methods:

Later particle content will be determined in residues of solubilized liver, lung, spleen, kidney, mesenteric and popliteal lymph nodes, Peyer's patches, and remainder of small intestine. Particles will be counted in appropriate aliquots of resuspended tissue residue or in residue deposited on small filters. Latex uptake and distribution will be determined in groups of mice given latex for 60 days and sacrificed at intervals up to 10 weeks after the termination of gavaging. Retention and distribution of ^{7}Be-labeled carbon will be determined by scintillation counting of organs from 0 to 336 hours after a dose of 10^{7} cpm.

Expected Results

The project will provide qualitative and quantitative information on the barrier function of the gastrointestinal tract and its changes with age. By determining particle uptake and distribution, factors which influence particle absorption will be elucidated and information on the mode of transport of particles away from the intestine will be gained. By determining particle retention and redistribution in intestinal and reticuloendothelial tissues, the project will provide information on the risks associated with the ingestion of environmental particulate pollutants in drinking water.

Title:
Direct Stochastic Model for Reactive Plumes

Principal Investigators:
P. Michael and
A. Levin

Institution:
Brookhaven National
Laboratory

Project Period:
2 years

Budget:
(2 years) $130,000

EPA Project Number:
R 809556

The objective of this program is to investigate theoretically the evolution of chemically reactive plumes in the atmosphere and to produce a mathematical formulation capable of describing the main features of the statistics of pollutant concentration fluctuations in plumes in which chemical reaction is significant. To accomplish this, statistical models of both the entrainment process and the mechanism by which intermittent chemical structure is developed inside the plume will be developed.

The first task to be undertaken is to develop a stochastic model for the entrainment process. Near the stack, if the chemical rate is sufficiently rapid, the process of turbulent mixing will determine the reaction rate for those emitted species which react primarily with species in the ambient atmosphere. Even for slower reactions, the turbulent mixing rate will determine the effective reaction rate by controlling the rate of dilution of initial concentration levels in the plume. Using the assumption that turbulent motion with scales in the inertial subrange dominates the entrainment process, a stochastic model for the growth of volume of a puff will be developed and then puff behavior will be related to plume evolution in the usual manner. The result will be a Fokker-Plank-type equation for the probability density function (pdf) of the volume of the puff, with parameters obtained from the atmospheric turbulence at the level of the plume and from dimensional considerations. The description will use Markovian statist: and will provide a stochastic description of volume growth somewhat analogous to the random trajectory model description of single-point diffusion in a shear flow.

The second task will be to develop a mathematical description of the intermittency process. The technique will follow the same lines as described in the treatment of entrainment. In this case a stochastic process will be proposed which represents the fragmentation of an initial puff into subsegments. A crucial role will be played by inertial subrange motions straining off parcels of the initial puff at time and length scales appropriate to the initial size of the puff and the scales of turbulent motion which are significantly smaller than the current puff dimension.

This project is primarily within the Atmospheric Sciences Division of the Department of Energy and Environment of Brookhaven National Laboratory.

The mathematical derivations and physical approximations will be carried out primarily by Dr. Arnold Levin, staff scientist within the Atmospheric Sciences Division; Dr. Paul Michael, Project Manager and Head of the Atmospheric Sciences Division, will review all derivations and approximations. There will be frequent consultations with staff members at Brookhaven National Laboratory who have knowledge of atmospheric chemistry, meteorology, and mathematical methods.

All reports and papers to be published are cleared through the Department of Energy and Environment.

Title:
Pollutant Chemistry in Combustion Systems

Principal Investigators:
I. N. Tang and
J. H. Lee

Institution:
Brookhaven National
Laboratory

Project Period:
1 year

Budget:
(1 year) $112,576

EPA Project Number:
R 809490

The objective of the research project is to identify important chemical reactions that may lead to a reduced emission of pollutants from fossil fuel combustion systems. During the first year of the project, research efforts will be focused on absolute rate measurements for high-temperature NO reactions with hydrocarbon radicals CH and CH_2. Reactions are carried out in a discharge fast flow apparatus specifically designed and constructed for operation at temperatures up to 1100K. Both atomic resonance fluorescence and laser-induced fluorescence techniques are currently employed for the detection and continuous monitoring of atoms and free radicals present in the reaction systems. It is expected that kinetic measurements, made under pseudo-first-order conditions as a function of temperature, will provide information for assessing the importance of such reactions as a potential NO_x destruction mechanism under staged combustion conditions. The research is expected to continue into its second phase concentrating on reaction product identification.

Title:

Development and Verification of a Method for Calculating Vapor Pressures from Gas Chromatographic Retention Data

Principal Investigator:
Professor James N. Seiber

Institution:
University of California
Davis, CA 95616

Project Period:
2 years

Budget:
(2 years) $69,944

EPA Project Number:
R 810134

The objective of this proposal is to develop and verify a method for calculating ambient vapor pressures of organic compounds of low volatility from gas chromatographic (GC) retention data. The work will entail development of the GC technique and approaches to calculation, examining GC conditions, candidate reference compounds, and methods of calculation in the process. The techniques examined will be applied to a variety of chemicals, with verification of accuracy and precision by comparison of GC-derived vapor pressures with experimental values in the literature or those determined experimentally in the course of this project. Approximately 60 individual organochlorine, organophosphorus, polynuclear aromatic, N-methylcarbamate, phenoxy acid/ester, aniline, and phthalate compounds, and several mixtures, will be examined. The aim is to develop a method which is rapid, accurate, and precise to provide vapor pressures of pesticides and other toxic substances of regulatory concern, and to compile a consistent set of vapor pressure data for such chemicals for development of structure-vapor pressures correlations.

It may well prove feasible eventually to estimate P from molecular structure. Having values of P for several classes of compounds determined by a consistent method may facilitate development of new structure-P correlations, as well as expanding the data base for evaluating currently available approaches to estimating P *via* boiling point (Grain, 1981).

Title:
A Study of the Reaction of Hydroxyl Radicals with Pernitric Acid

Principal Investigator:
Mario J. Molina

Institution:
University of California, Irvine

Project Period:
1 year

Budget:
(1 year) $30,244

EPA Project Number:
R 809742

The objective of this research is to measure the rate constant for the reaction of hydroxyl radicals, OH, with pernitric acid vapor, $HOONO_2$. This rate constant value is required for model calculations of the chemistry of the stratosphere: our earlier results indicated that pernitric acid is very stable towards solar photolysis, and hence reaction with hydroxyl radicals is expected to be the predominant destruction process for this species in the lower stratosphere. Furthermore, the reaction is expected to have a pronounced effect on the hydroxyl radical balance and hence on the stability of stratospheric ozone towards anthropogenic perturbations resulting from the industrial release of chlorofluorocarbons.

We are measuring the absolute rate constant for the OH + $HOONO_2$ reaction by means of the flash photolysis resonance fluorescence technique. Pernitric acid is a rather unstable species in the laboratory and it cannot be prepared in pure form. Mixtures of $HOONO_2$ vapor with H_2O, H_2O_2, and HNO_3 are prepared by flowing He, a carrier gas, through concentrated aqueous solutions containing H_2O_2 and HNO_3 vapors. The composition of the gaseous mixture is established by long-path Fourier-transform infrared spectroscopy, and the measured pseudo-first order OH decay rates are corrected for the contributions due to H_2O_2 and HNO_3. Our preliminary results indicate that these corrections amount to at most 30% of the overall rate, and that the rate constant value at room temperature is about 5×10^{-12} molecule cm^{-3} s^{-1}, a value considerably larger than the one originally used in model calculations. Furthermore, our preliminary results yield a negative activation energy, with a value of about 1.2 kcal/mole, which is comparable to the value for the reaction of OH with HNO_3.

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Title:

Scavenging of Gases and Aerosol Particles by Atmospheric Clouds and Precipitation and its Contribution to Acid Precipitation

Principal Investigator:
Hans R. Pruppacher

Institution:
University of
California
Los Angeles, CA

Project Period:
2 years

Budget:
(2 years) $206,589

EPA Project Number:
R 809335

The present study attempts to get a better understanding of the processes which control the scavenging of atmospheric trace gases by clouds and precipitation. In particular the two gases, Sulfurdioxide (SO_2) and Nitrogendioxide (NO_2), responsible for what is known as "acid rain" are being studied. A theoretical model and sophisticated experimental set up, developed and perfected during previous EPA grants, are used in this study. The theoretical model involves a numerical solution of the convective diffusion equation for a trace gas diffusing through the environment toward—and into—falling water drops inside of which the water is exhibiting a recirculating flow. The experimental set up involves a 35 meter high shaft adapted for studying the efficiency with which water drops of various sizes are able to remove a gas present in various concentrations. Both the rate of gas-absorption and gas-desorption are studied. The results obtained are used to determine the rate at which a gas contained in a pollution layer of specified extent is removed by a rainfall of specified precipitation intensity and drop size distribution.

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Title:
Dose Response Curves for Multiple Trace Elements

Principal Investigator:
Arthur Wallace

Institution:
University of California
Los Angeles, CA 90024

Project Period:
2 years

Budget:
(2 years) $95,533

EPA Project Number:
R 809578

Trace element toxicity is an environmental problem and environmental impact statements are often required to contain predictions as to whether or not a given technology will result in trace element toxicity to plants, animals or man. The criteria employed for assessing possible toxicity are often in considerable error because they do not take into account possible additive and synergistic effects when two or more elements are simultaneously near the toxicity range. Accurate assessment demands that these phenomena also be considered. An example of the powerful influence that additive effects can have is that if enough of each of 22 different trace elements were present in sufficient excess to each result in only a 5% yield decrease if contaminating separately, all contaminating together can result in a yield decrease to 32% of maximum if each were additive. The effect can be even more drastic if synergistic effects are involved.

The objectives of the study are:

1. Refine the data base for threshold toxicity levels of trace elements through consideration of interactions present when 2, 3, and 4 different trace elements are in excess simultaneously (this causes variation in threshold levels, lethal levels and in the amount of toxicity per unit of metal).
2. Better develop the theoretical basis for explanation of multitrace element interactions in plants which modify phytotoxicities.
3. Prepare a rational scenario, which could guide those making environmental impact statements, on potential toxicities when more than one trace element is involved in pollution insofar as our studies with four elements simultaneously will permit.

Studies to date are indicating both additive and synergistic effects when two and three trace elements are contaminating simultaneously. Equally important is that threshold levels are being altered with evidence that a critical level for a given trace element is lower when other trace elements simultaneously contaminate. From the studies more realistic understanding of critical levels is expected.

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Title:

Investigation of Limiting Engineering and Chemical Factors for Recycle and Reuse of Blast Furnace Scrubber Waters under the Clean Water Act and RCRA

Principal Investigator:
Richard G. Luthy
Francis C. McMichael
and Mitchell J. Small

Institution:
Carnegie-Mellon University

Project Period:
2 years

Budget:
(2 years) $163,134

EPA Project Number:
R 809628

The purpose of this study is to develop an integrated approach to the recycling, treatment and management of blast furnace scrubber water. The study will assess the recycle of blast furnace water in scrubber and spray towers and the treatment of recycled water to achieve coincident removal of recycle-limiting and environmental related constituents, with disposal of a minimized blow-down stream by slag quenching. Issues to be studied in this investigation include: (1) evaluation of quality constraints imposed on the streams for which the treated water may be reused; (2) investigation of physico-chemical phenomena which influence the behavior and fate of process (recycle-limiting) contaminants and environmental contaminants; and (3) evaluation of the effect upon slag leachate characteristics of quenching slag with waters containing varying levels of toxic metals and organics. The study includes bench-scale testing with scrubber waters, and chemical and process modeling to predict water quality changes and precipitate formation upon reuse and treatment. The results of this study will provide a fundamental understanding of the fates of recycle-limiting species and environmental contaminants in industrial wastewater reuse systems, and provide guidance for the improved operation of blast-furnace recycle systems.

Title:

Chemical Basis of the Mutagenicity and Potential Carcinogenicity of Environmental Agents

Principal Investigator:
Gilles Klopman and Herbert S. Rosenkranz

Institution:
Case Western Reserve University

Project Period:
2 years

Budget:
(2 years) $146,121

EPA Project Number:
R 810063

In this project an attempt will be made to predict the mutagenic and toxicological properties of chemicals by establishing quantitative structure activity relationships. The chemical structural features of environmental chemical agents will be evaluated as a function of their respective biological activities. The approach employs definition of the physiochemical features of mutagenic sets of compounds based on molecular mechanics, MM, quantum mechanics, QM, and pattern recognition techniques, PR.

Both linear free energy (LFE) and non-linear free energy (NLFE) QSAR will be investigated. In the case of NLFE, action models will be proposed to explain the experimental data. The initial data set considered will be composed of the families of compounds whose mutagenicity was tested in the Gene-Tox program, and that are currently being compiled by the Center for the Environmental Health Sciences of Case Western Reserve University.

Within each series of congeneric chemicals, the determination of the relationship of mutagenic potency to chemical structure will be undertaken for every assay for which sufficient experimental results are available. Common endpoint screens will be developed to provide biological activity for the QSAR analysis. The QSAR which emerges from this work will be tested by choosing additional compounds from the data base. Whenever necessary, we will also augment the data base by synthesizing and testing chemicals whose mutagenicity determination may help resolve ambiguous mechanistic paths suggested by the action models.

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Title:

Molecular Biology and Usefulness of Nitroreductase-less Bacteria

Principal Investigator:
Herbert S.
Rosenkranz

Institution:
Case Western
Reserve University

Project Period:
2 years

Budget:
(2 years) $87,000

EPA Project Number:
R 809881

Recently it was recognized that nitropyrenes as well as other nitrated polycyclic aromatic hydrocarbons (nitroarenes) are widely distributed in the environment chiefly as a result of the replacement of the gasoline-powered passenger car by diesel-driven automobiles. This dieselization trend is projected to continue. Because nitropyrenes as well as other nitroarenes are highly mutagenic in a number of systems and as some of them have been reported to be cancer-causing as well, it is of importance to quantitate their presence in the environment. The traditional techniques of the chemist (fractionation and instrumental identification of the chemicals) do not lend themselves routinely to this task because of their complexity, the requirement for expensive equipment and trained personnel. It is proposed to take advantage of the potent mutagenicity of these chemicals in microbial systems to construct bacterial strains that are specifically deficient in their response to the genetic action of various members of this group of chemicals.

This recently initiated project has resulted, thus far in the following:

1. A strategy has been devised that is useful and effective in constructing a variety of such bacterial mutants.
2. A collection of such strains has been obtained and their characterization has been initiated.
3. Initial results indicate that such strains are useful for analytical purposes, especially with respect to complex mixtures.
4. These preliminary studies have revealed that bacteria possess a diversity of enzymes that are involved in the bioconversion not only of nitroarenes but of other metabolites of environmental interest, such as the aromatic amines as well.
5. The most unexpected finding of these preliminary studies has been the discovery of a, heretofore unsuspected enzyme which specifically esterifies arylhydroxylamines. The role of this enzyme the mutagenicity of aromatic amines as well as of nitroarenes and its possible presence in mammalian cells will be a major thrust of future studies as it is, presumably, also involved in the carcinogenic process.

Title:
Nitropyrenes: Do They Constitute an Environmental Hazard for Man?

Principal Investigator:
Herbert S.
Rosenkranz

Institution:
Case Western
Reserve University

Project Period:
3 years

Budget:
(3 years) $342,301

EPA Project Number:
R 809476

Recently it was recognized that nitropyrenes as well as other nitrated polycyclic aromatic hydrocarbons (nitroarenes) are widely distributed in the environment chiefly as a result of the replacement of the gasoline-powered passenger car by diesel-driven automobiles. This dieselization trend is projected to continue. Because nitropyrenes as well as other nitroarenes are highly mutagenic in a number of systems and as some of them have been reported to be cancer-causing as well, it is of importance to determine the biochemical basis of these varied mechanisms of action. This project addresses the problem at the cellular and organismic level. The biotransformation, reaction of the metabolites with cellular DNA and the repair of the affected DNA is being investigated.

This recently initiated project has already shown that there is a basic dichotomy in the biotransformation of some nitroarenes in bacterial and mammalian systems. This may account for the extraordinarily potent activity of these chemicals in microbial systems.

A matrix was developed to extract additional information from bacterial mutagenicity assays performed in the presence as well as the absence of mammalian enzymes such as to obtain clues regarding mechanisms. These can then be followed with specific studies. Indeed, using the nitroarene 5-nitroacenaphthene the value of this method has been proven as it demonstrated the existence of additional pathways.

Recently, a new form of DNA, the Z-DNA, has been recognized. The role of this newly discovered DNA in cancer-causation is, as yet, not established. It is known, however, that some cancer-causing chemicals, including the bioconversion products of the nitroarenes may react with Z-DNA to yield unusual structures. In addition, the reaction of such metabolites with "normal" DNA may also induce it to assume the Z-configuration. In an attempt to evaluate the role of this DNA with respect to the induction of mutations and cancer, we have attempted to develop a predictive assay that will enable us to determine the role of Z-DNA in biological phenomena. Our preliminary results indicate that we may have succeeded in developing a rapid as well as a simple assay capable of ascertaining whether or not an environmental chemical or its metabolites may induce the Z-DNA conformation. This will enable us to screen for such chemicals in the environment.

Title:

Investigation of the Transport and Diffusion of Pollutants in Complex Terrain Using a Numerical Model

Principal Investigator:
Roger A. Pielke

Institution:
Colorado State University

Project Period:
1 year

Budget:
(1 year) $59,606

EPA Project Number:
R 810471

The overall objective of this research is to utilize a primitive equation mesoscale model in order to simulate atmospheric flow patterns induced by complex terrain, and to examine the impact of these flow fields on the transport and diffusion of air pollutants. By comparing the mesoscale model results with available observations from previous and current field experiments (including those of EPA in complex terrain), we are quantitatively establishing the degree of accuracy of the model.

Our work utilizes the mesoscale model predicted winds and turbulence characteristics in order to provide input to a semi-stochastic transport and diffusion simulator. Also called the Lagrangian conditioned particle scheme, this approach permits the estimation of the spread of a pollutant from a point source in a region in which there are substantial temporal and spatial variations in the flow field. Using new observational information on turbulent scales and boundary layer structure in complex terrain, we also expect to improve and refine this parameterization of the spread of plumes and puffs.

Our modelling experiments are performed on both idealized and real terrain configurations. The simpler, ideal topography provides the needed insight into the physical mechanisms involved with flow in complex terrain, while simulations of actual geographic regions establishes the quantitative limits of our model.

A major practical result of our studies will be the establishment of a tool with which to estimate the pollution concentrations in irregular terrain which result from specific sources of air pollution. A substantial improvement to the current operational procedures, the use of a mesoscale model, will also provide the scientific explanation for the mechanisms of formation and evolution of flows which develop in complex terrain.

Title:
New Fuel Additives for Reducing Pollutants from Combustion

Principal Investigator:
Prof. Robert E. Sievers

Institution:
University of Colorado

Project Period:
2 years

Budget:
(2 years) $272,099

EPA Project Number:
R 808312

This research program is a study of the effects of novel fuel additives on gaseous exhaust emissions. These investigations involve the synthesis and characterization of new lanthanide and transition metal chelates and a study of the effects of these compounds when dissolved in fuels on the formation and emission of carbon monoxide (CO), hydrocarbons (HC), and oxides of nitrogen (NO_x) from internal combustion engines. Experimentally, exhaust from a test engine will be monitored with infrared, chemiluminescent, gas chromatographic, and mass spectrometric instrumentation, which will allow sensitive and selective evaluation of additive effects on exhaust gas emissions. Such data may lead to new insights and understanding of the manner in which combustion is modified in the presence of metal species. In addition, a study of a novel means of regenerating poisoned catalytic converters by formation of noble metal films during combustion will be made. The expected result is knowledge of whether certain novel fuel-soluble additives will reduce noxious emissions from combustion sources.

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Title:

The Destruction of Hazardous Organic Substances in a Slagging Gasifier Environment and the Encapsulation of Toxic Heavy Metal Compounds in a Nonleachable Vitreous Matrix

Principal Investigator:
Helmut W. Schulz

Institution:
Columbia University

Project Period:
15 months

Budget:
(15 months)
$269,230

EPA Project Number:
R 808965

This project expects to determine the technical feasibility of utilizing the Simplex gasification process for hazardous waste disposal. The Simplex process produces a clean, medium heating value fuel gas from a briquetted feed of coal and municipal wastes, without generating stack emissions or contaminated residues. The process has the potential for disposing of a wide variety of toxic wastes, including halogenated hydrocarbons and heavy metals.

The project addresses two separate, but interrelated, research investigations which are superimposed on a fully developed process for the slagging gasification of a briquetted feedstock comprising Eastern bituminous coal and cellulosic waste materials (such as urban waste or forest debris).

It is proposed to demonstrate hazardous waste destruction by the pulsed addition of toxic wastes into the prototype Simplex gasifier. Experimental work will address two waste types which are treated separately in this proposal: Highly refractory analogs of PCBs and other toxic organics (Part A) and industrial sludges containing heavy metals (Part B).

Part A of the proposed research will provide quantitative data on the completeness of destruction, at varying throughput rates, of such refractory analogs as hexachlorobenzene. These substances will be introduced into the hearth of a slagging gasifier where they encounter the pyrolytic environment of incandescent char at a temperature of 3000 degrees F in the presence of steam and pure oxygen. The residence time at temperatures above 2000 degrees F is believed to be more than adequate to ensure complete thermal degradation in a single pass.

Part B calls for a quantitative determination of the degree to which heavy metal compounds present in industrial or sanitary sludges are captured and encapsulated in the vitreous slag tapped from the hearth of the gasifier. To the extent that small concentrations of heavy metals are carried over with the crude product gas, they are condensed and recovered with the tars and recycled to the hearth of the gasifier. Part B also includes a determination of the comparative resistance to leaching by ground water of the heavy metal compounds encapsulated in the fritted slag.

In addition to providing the quantitative analytical data just described, the study will demonstrate the technical feasibility of employing the Simplex and biomass gasification process as a cost-effective vehicle for the disposal of toxic waste materials.

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Title:
Anaerobic Digestion of Waste Activated Sludge

Principal Investigator:
James M. Gossett

Institution:
Cornell University

Project Period:
2 years

Budget:
(2 years) $141,376

EPA Project Number:
R 809500

In the literature, there are numerous reports concerning the anaerobic digestion of waste activated sludge (WAS). However, no comprehensive models exist which allow the prediction of digester performance as a function of activated sludge process parameters. Based upon previous studies by the Principal Investigator, a model has been proposed which attempts to predict: (1) ultimate anaerobic digestibility of WAS as a function of activated sludge influent and process parameters (such as solids retention time); and (2) anaerobic digester performance as a function of ultimate digestibility and digestion parameters (such as solids retention time in the anaerobic process). The ultimate digestibility portion of the model has been previously validated. However, the portion concerning kinetics has not been rigorously tested. The model postulates that membrane-protected viable WAS cells become available substrate to anaerobic organisms at a rate which is first-order with respect to the remaining concentration of viable cells in the digester.

The purpose of this investigation is to experimentally evaluate the model. Semi-continuous digestion studies at a variety of retention times and influent concentrations will be performed, using WAS which has been generated in the laboratory. The effects of preliminary disruption of cell membranes upon digestion kinetics will be evaluated. Batch studies examining the rate of WAS solubilization in the presence of various concentrations of digester sludge will also be part of this investigation.

Thus far, eight laboratory-scale digesters have been set up and are operating on a WAS feed (laboratory generated). These digesters are continuously-mixed and fed hourly (approximating continuous flow conditions). Two solids feed concentrations are being employed, and four solids retention times. Steady-state conditions have not yet been achieved.

Title:

Qualitative Environmental Impact Assessment: Marine Plankton Communities

Principal Investigator:
Patricia A. Lane

Institution:
Dalhousie University
Halifax, Nova Scotia

Project Period:
2 years

Budget:
(2 years) $93,254

EPA Project Number:
R 810520

The objectives of this study are: 1) to apply a qualitative ecosystem analysis to a set of field and laboratory marine plankton communities; 2) to evaluate the qualitative models with a set of new community structure-stability descriptors; 3) to compare all communities with these descriptors to identify the underlying core structure; 4) to generate all of the necessary computer software to handle the qualitative techniques; and 5) to study and develop model verification procedures and selection of the "best fit" model. The overall research goal is to develop a new way of characterizing natural communities that has predictive capabilities in terms of evaluating the cause-effect pathways of stresses to ecological systems.

The research approach centers on loop analysis, a qualitative network technique, that facilitates the identification of ecosystem structure and functional relationships. To date, five ecosystems have been modelled and an underlying core structure has been identified. We have been able to document shifts in system sensitivity to stress in ecosystems undergoing nutrient enrichment. We can outline the causal pathways between a stress to these networks and all variables within the network.

Expected results from this study include a rigorous testing of loop analysis as a tool for enhancing understanding of ecosystems undergoing stress and preliminary application of this tool for improving current methods of environmental impact assessment. Present assessment methods include costly computer simulations which are strongly quantitative and require large detailed data sets. Loop analysis could reduce the amounts of data needed with a concurrent increase in understanding and a substantial decrease in overall assessment costs. Thus, instead of measuring everything, loop analysis would indicate what is really necessary to measure in a complex ecological system.

Title:
Teratogenic Evaluation of Chronic Heavy Metal Exposure

Principal Investigator:
Vergil H. Ferm

Institution:
Dartmouth Medical School,
Hanover, NH 03755

Project Period:
3 years

Budget:
(2 years) $183,679

EPA Project Number:
R 810078

This study will evaluate a method for the *in vivo* study of chronic-subacute exposure of the pregnant animal to certain heavy metals and arsenate, all of which are potentially significant environmental threats. The metals to be studied have well-defined teratogenic endpoints in our animal model, following acute bolus doses. To study chronic-subacute exposure, we will implant osmotic minipumps into the pregnant hamsters during the early states of gestation. This method will assure a sustained and consistent supply of the teratogen to the maternal and embryonic system during the early period of pregnancy—a period which corresponds to the first 30 days of human gestation—and the time during which the embryo is most susceptible to teratogenic agents. This technique might well have several advantages over acute bolus or *in vitro* techniques currently being used for teratogenic testing. It certainly is more realistic as a method of exposure to environmental teratogens than are the other methods. It retains the normal physiological status of an intact materno-placental-embryonic unit. Both the amount and length of exposure to these molecular teratogens can be accurately controlled. The second part of our project proposes to explore certain aspects of the biochemical state of each of the selected teratogens in the materno-embryonic unit during teratogenesis. We are particularly interested in determining the degree to which the concentration and chemical status of each teratogen in specific tissues of the maternal and embryonic systems correlates with the level of teratogenic response. This information could help pinpoint the site (or sites) of teratogenic insult and could lead to an explanation of the mechanism of heavy metal-induced teratogenesis. These studies should determine whether or not this method offers a more accurate and realistic measure of the teratogenic threat of environmentally significant heavy metals than current bolus type studies.

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Title:
Oxidation of Cyanides in Industrial Wastewaters by Ozone: Kinetics and Mechanism

Principal Investigator:
Dr. Mirat D. Gurol

Institution:
Drexel University

Project Period:
2 years

Budget:
(2 years) $139,900

EPA Project Number:
R 809613

Wastewaters generated by various industrial activities contain substantial quantities of cyanide compounds whose discharge into environment might create serious ecological problems. Ozone is capable of oxidizing these compounds.

The primary objective of this research is to generate the fundamental kinetic and mechanistic data about the ozonation of 1) simple cyanides, 2) the reaction intermediate, cyanate, and 3) the complexes of cyanide with various metals, i.e., Cd, Zn, Cu, Ni, Fe. The experiments are performed in batch reactors under quiescent conditions. A Stop Flow Analyzer is used to study the kinetics of the fast reactions. Attention is given to distinguishing between the oxidation by molecular ozone, and the oxidation by free radicals arising as decomposition products of ozone. It is also the objective to develop dynamic models describing the overall rate of cyanide removal in continuous flow ozone reactors where the kinetics of mass transfer of ozone become relevant. The data produced in batch reactors will be incorporated into the models. The models will be tested by comparing their predictions for cyanide, cyanate, and ozone concentrations in the effluent, and for the ozone consumption with the measurements obtained under various experimental conditions in a Continuous Flow Stirred Tank Reactor.

It is expected that the results of this study will provide the engineers with the essential tools for designing and operating the ozone reactors, and will contribute to the literature on mechanisms of ozonation reactions.

Title:
Role of Lipid Peroxidation in Air Pollutant Toxicity

Principal Investigator:
Daniel B. Menzel

Institution:
Duke University
Medical Center

Project Period:
2 years

Budget:
(2 years) $172,000

EPA Project Number:
R 809283

The proposed studies are directed to evaluate the role of lipid peroxidation in O_3 and NO_2 toxicity. An initial objective is to test the effect of incorporating specific unsaturated fatty acids into lung lipids on the toxicity of O_3 and NO_2, and to relate this toxicity to the theoretical formation of prostaglandin-like endoperoxides. Only fatty acids having 3 or more ethylenic groups are capable of forming endoperoxides. If the toxic effect of lipid peroxidation initiated by O_3 and NO_2 is caused by endoperoxides, then the feeding of diets which increase lung monoenoic or dienoic fatty acids should have little effect on the toxicity, whereas a diet which increases the trienoic and polyenoic fatty acid content should exacerbate toxicity. These studies will evaluate the effect of the type of lung fatty acid unsaturation rather than the extent of unsaturation alone (peroxidizability index) on oxidant injury.

A second objective is to evaluate the effects of O_3 and NO_2 exposure on the kinetics of vitamin E uptake and turnover in the lung. Vitamin E is the major dietary factor affecting the toxicity of O_3 to mice and rats and of NO_2 to rats. Changes in lung fatty acid composition observed after O_3 exposure may be a reflection of changes in the tissue vitamin E levels. Vitamin E and its metabolites will be determined in the lungs of mice fed defined diets and exposed to O_3 or NO_2.

A third objective is to correlate these effects with those occurring in man. Because there is a need to extrapolate animal exposures to humans, simple tests of oxidant injury applicable to man will be investigated. RBC filterability has been used as an index of injury for other toxicants and may be useful to quantitate O_3 and NO_2 injury to mice fed defined diets. The exhalation of ethane and pentane is also easily adapted to man and will be used to estimate O_3 and NO_2 catalyzed peroxidation *in vivo* and its prevention by vitamin E.

These studies will provide direction for human studies and, together with the literature on other effects of O_3 and NO_2, will aid EPA in fulfilling its role in setting standards.

Title:

Corona Wind and Turbulence in Full Scale Electrostatic Precipitators

Principal Investigator:
Edward J.
Shaughnessy

Institution:
Duke University

Project Period:
2 years

Budget:
(2 years) $189,713

EPA Project Number:
R 809727

The production of turbulence in full scale electrostatic precipitators by corona wind will be investigated using a new large aspect ratio wire to plate model precipitator. Hot-wire anemometer surveys of the turbulence at the inlet and exit of the precipitator section will be obtained at various air speeds and current densities under particle free, cold gas conditions. Mean velocity profiles, mean square velocity fluctuation profiles, and the power spectra of the turbulence will be examined for evidence of turbulence production by the electrical body force. The model precipitator will have an aspect ratio of 20 at a 10 inch plate to plate spacing, and will operate at flowspeeds of 1 to 10 ft/sec. The aerodynamic turbulence of the facility will be characterized by hot-wire surveys with the current off. These results will be compared with profiles taken with the corona current at various current densities spanning the commercial range of interest. Both positive and negative corona will be employed. The primary questions of interest are whether there is significant production of turbulence due solely to the corona current, and if so, how this production mechanism varies with flow speed, current density, and polarity.

Title:
Fate of Viruses and Trace Organics in a Cypress Strand Treated with Chlorinated Primary Effluent at Waldo, Florida

Principal Investigator:
Gabriel Bitton and
Samuel R. Farrah

Institution:
University of Florida

Project Period:
2 years

Budget:
(2 years) $195,000

EPA Project Number:
R 809402

This research proposal deals with the public health aspects of sewage disposal in wetlands, particularly cypress strands which are "slow flowing" wetlands. A cypress strand in Waldo, Florida, has been receiving chlorinated primary effluents for several years. We need to know more about the survival pattern of bacterial and viral pathogens as well as the formation of mutagenic compounds in these highly colored (with humic/fulvic substances) waters.

During the first year of this project we will attempt to develop the methodology for virus recovery from these highly colored waters and from the underlying sediments. We will also determine the decay rates of laboratory-grown viruses, bacteria and phages in survival chambers under *in situ* conditions in the cypress strands. In addition, two experimental corridors have been constructed in the swamp and will be used to determine the capacity of the strand to remove bacteria and viruses. Sewage will be pumped into these corridors, and viruses, phages and bacterial indicators will be monitored before and after their passage through the corridors.

The second year will be devoted to monthly monitoring of indigenous viruses in the cypress strand to determine the potential of these low-energy systems to remove viruses. We will also monitor trace organics in the chlorinated primary effluents, as well as in sediments and water in the cypress strand. The mutagenicity and identity of the isolated trace organics will be determined using short-term bacterial assays and GC-MS, respectively. If mutagenic chlorinated compounds are found we will experimentally determine the potential of fulvic/humic substances present in strand water to form mutagenic substances under conditions prevailing in the cypress strand.

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Title:
Electrostatic and Hydrophobic Interactions in Virus Adsorption to Solids

Principal Investigator:
Dr. Samuel R. Farrah

Institution:
University of
Florida

Project Period:
2 years

Budget:
(2 years) $149,299

EPA Project Number:
R 810126

The association of viruses with solids such as membrane filters, soils, sediments, and wastewater sludges has been the subject of several previous investigations. These studies have shown the importance of electrostatic interactions in determining virus adsorption to solids. More recently, the contribution of hydrophobic interactions to virus adsorption to solids has been investigated. The main objective of this study will be to determine the relative contributions of both electrostatic and hydrophobic interactions in the adsorption of different viruses to a variety of solids. By studying the factors involved in virus adsorption, we hope to be better able to explain certain results such as the reported differences in the ability of viruses to adsorb to a particular solid such as soil. Also, we would like to explain the differences in the abilities of different solids to adsorb a particular virus under different conditions. In most cases, viral adsorption to solids will be studied by measuring the level of virus adsorbed to a solid. Viruses will be measured as infectious plaque-forming units. The effect of solutions that either promote or interfere with electrostatic or hydrophobic interactions on the extent of virus adsorption will be examined. Results of this study should help in determining the relative strengths of the forces maintaining virus adsorption to solids under different conditions. This will permit a better understanding of the factors that influence the distribution of viruses in the environment. By better understanding the factors influencing virus adsorption to and elution from solids, we hope to develop more efficient and simpler procedures for recovering viruses from environmental samples. In summary, the basic factors involved in virus adsorption to solids will be studied. The results should permit a better understanding of virus distribution in the environment and permit development of better procedures for detecting viral contamination in the environment.

Title:
Vegetation Development in an Alaskan Salt Marsh

Principal Investigator:
Susan W. Vince

Institution:
Center for Wetlands
University of Florida

Project Period:
2 years

Budget:
(2 years) $139,869

EPA Project Number:
R 809874

Alaskan salt marshes have received little ecological study despite their extent, productivity, and importance for waterfowl populations. The proposed research seeks to identify patterns of vegetation development and to determine the mechanisms governing these processes on Susitna Flats, a marsh on the south-central Alaska coast. Three types of techniques—quantitative plant and environmental analysis, laboratory experiments, and controlled field manipulations—will be employed. Plant species composition and production, tidal exposure, sedimentation rate, and substrate characteristics will be measured at a variety of sites on Susitna Flats, providing a description of successional processes. Whether plant zonation is in fact homologous with temporal succession will be determined by examination of the vertical accretion of plant debris in the sediment, by comparison of the vegetation of sites of differing ages, and by observation of long-term vegetation changes in marked plots. Laboratory and field experiments are designed to test hypotheses concerning the reasons for the delay in appearance of most plant species on new sites, and the disappearance in time of the colonizers. The specific and combined effects on plant growth of presumed driving forces—soil type, salinity, waterlogging, and nutrient availability—will be determined. It is anticipated that this experimental approach will enable predictions of plant community response to both natural (e.g. earthquakes) and human disturbances.

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Title:
Oxidative Destruction of Organic Groundwater Contaminants

Principal Investigator:
James N. Butler

Institution:
Harvard University,
Division of
Applied Sciences

Project Period:
2 years

Budget:
(2 years) $142,350

EPA Project Number:
808209

Contamination of groundwater by toxic chemicals has led to abandonment of water supplies, or to costly installation of treatment systems based on activated carbon. The objective of this project is to investigate methods for oxidative destruction of these contaminants. Methods will include ozone, ozone together with ultraviolet irradiation, and singlet oxygen generated by reaction between hypochlorite and hydrogen peroxide.

Experimental research will consist of measuring the extent of reaction and nature of products for a selected list of compounds (e.g., chloroform, dichloroethylene, trichloroethylene, tetrachloroethylene, dichloroethane, trichloroethane, vinyl chloride, chlorobenzene, etc.), all of which have been found as pollutants in groundwater supplies. Primary variables will include reagent concentration, pH, temperature, etc. The reactions will be followed by gas chromatography with confirmation of peaks by gas chromatography/mass spectrometry. Special efforts will be made to identify reaction mechanisms and interferences characteristic of the ppm level in fresh water.

Results of this investigation should make it possible to design treatment systems for a groundwater supply known to be polluted by a particular set of chemicals, and to optimize the cost of such treatment.

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Title:
Toxicity of Priority Pollutants on Tropical Oceanic Plankton

Principal Investigator:
Jed Hirota

Institution:
University of
Hawaii, Manoa

Project Period:
2 years

Budget:
(2 years) $326,882

EPA Project Number:
R 809916

Our project funded by EPA has two major objectives: 1) develop new methods and techniques to enable the careful, live collection, handling, and laboratory cultivation of tropical, oceanic plankton for their entire life cycle from egg back to egg; and 2) evaluate the effects of halomethane and haloamine chemical species, which result from the chlorination of seawater, on these selected plankton species at sublethal, chronic exposures. The research plan is to begin with gentle collection methods and cultivation techniques first, and then to proceed on with the toxicity testing and "bioassays" after the new techniques and feeding conditions are determined.

The expected results from this project include new information on the methods, techniques and data required for the successful cultivation of these tropic oceanic plankters over their entire life cycles (e.g., their rates of growth, reproduction and feeding). These data are not currently available for virtually all of the commonly occurring oceanic species of plankton in the tropical regions. Secondly, we expect to develop some new bioassay measures to evaluate subtle changes to animal growth, reproduction and feeding rates in response to sublethal, chronic exposures to the major byproducts of chlorination of seawater.

It is anticipated that this new information will be useful and applicable to an evaluation of effects of low levels of chlorination of seawater (as a biocide and anti-fouling agent) for future OTEC (Ocean Thermal Energy Conversion) power plants and also for existing wastewater or conventional power generating plants. Our studies are being geared toward an assessment of the potential effects of seawater chlorination in the far-field plumes of discharges of any type on tropical oceanic plankters.

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Title:
Scintigraphic Studies of the Pharmacokinetics of Delayed Neurotoxins

Principal Investigator:
Larry G. Hansen

Institution:
University of Illinois

Project Period:
2 years

Budget:
(2 years) $114,904

EPA Project Number:
R 809845

The summary objectives of the project are to define the early movement of organophosphorus "delayed neurotoxins" within the laying hen. The hen is the standard model animal because 1) many other species are resistant to these effects and 2) the effects are irreversible so that human studies cannot be conducted. The compounds differ from many other similar pesticides in certain chemical properties, but very similar compounds may have very dissimilar potencies. Compound access to the central nervous system shortly after exposure appears to determine the severity of ataxia and/or paralysis several days later. Methods are to be developed and used to evaluate this early distribution which is, to date, very difficult to monitor in the laying hen. Potent and less potent analogs will be compared.

Methods will include gamma counting and gamma imaging with ^{125}I analogs as well as traditional ^{14}C tracer studies. The initial phase is to synthesize active analogs containing iodine (activity will be tested in hens). It is anticipated that differences in absorption, disposition and biotransformation will account, at least in part, for divergent potencies in closely related analogs.

Title:
Role of Surface Active Media in Expanded-Bed Anaerobic Filters

Principal Investigator:
Makram T. Suidan
and Bruce E.
Rittmann

Institution:
University of
Illinois at
Urbana-Champaign

Project Period:
3 years

Budget:
(2 years) $167,355

EPA Project Number:
R 809750

The overall objective of this research project is to determine the effect of surface activity of granular activated carbon on biofilm accumulation characteristics in completely mixed, expanded-bed anaerobic filters. The research will test the hypothesis that superior biofilm accumulation allows lower effluent concentrations and will determine the cause of better attachment. In addition, the research will develop a fundamental kinetic model that will be used to design the most fruitful experiments for evaluating the important concepts and determining the operational limits of the process. Granular activated carbon represents a surface-active-attachment medium, while crushed anthracite coal, of an equal mesh size, represents a non-surface-active medium. Biofilm accumulation and substrate utilization characteristics for various loadings of two substrates, acetate and phenol, will be evaluated. Acetate is selected in order to provide knowledge of biofilm attachment and utilization kinetics for the sensitive and slow-growing methanogenic organisms. Phenol, on the other hand, represents a substrate that would result in the growth of mixed cultures of methanogenic and nonmethanogenic organisms. The strong adsorption characteristics of phenol will also permit the evaluation of the role of substrate adsorption by activated carbon on the attachment and accumulation of methanogens.

The anaerobic filter performance will be modeled using a mathematical representation that includes substrate transport and utilization, biomass growth and decay, and biomass attrition due to hydrodynamic shear forces. The mathematical model will be used in selecting experimental loading conditions that will optimize the evaluation of reactor performance over a wide range of process conditions.

Title:
Effects of Microwave Exposure on the Mammalian Immune System

Principal Investigator:
Wayne A. Tompkins

Institution:
University of Illinois
Urbana, IL 61801

Project Period:
3 years

Budget:
(2 years) $323,328

EPA Project Number:
R 809566

The principal objectives of the proposed research are to determine the effects of Microwave (MW) exposure on the immune response in mammals and to determine quantitative relationships between threshold and super-threshold exposure parameters and observed immunological alterations. This proposal will investigate the effect of acute high dose and chronic low dose exposure of MW radiation on the mammalian immune system using several mouse and hamster test systems.

These objectives will be attained by studying the effects of MW on hamster and mice natural immunity and their ability to respond immunologically to a) vacinnia virus, b) synthetic T-dependent and T-independent antigens, and c) mitogens. Well defined *in vitro* assay systems, to test for MW induced immunodepression or immune stimulation, will be used studying a) antibody responses, b) T cell responses, c) macrophage, and d) natural killer activity.

The immune systems comprising of complex network of interacting cells involving helper, suppressor and effector functions is believed to be extremely sensitive to MW induced perturbations. MW exposure has been shown to cause immune suppression and other system changes. If MW at same intensity and exposure duration have immunosuppressive effects, the greatest risk of such exposure might represent increased susceptibility to virus infections. From the study on different parameters of immunity to vaccinia virus and other antigens, we will be able to make precise quantitative determinations of intensity and duration of MW exposure required to produce immune system changes. This information will be of major significance in determining MW exposure safety standards.

Title:

Fates of Organic Compounds from Niagara Falls Dumpsites in Lake Ontario

Principal Investigator:
Dr. Ronald A. Hites

Institution:
Indiana University

Project Period:
3 years

Budget:
(2 years) $115,422

EPA Project Number:
R 808961

Objectives:

Toxic organics have been leached and are being leached from dumpsites in Niagara Falls into the Niagara River and into Lake Ontario. The major objective of this project is to determine the long-term fates of those compounds which enter Lake Ontario in this way. Ten to twenty compounds, which have already been identified, will be investigated in lake sediment and biota. A considerable effort will be spent mapping the fluxes to the sediment of these compounds. Flux measurements will allow intercomparison of data and the calculation of mass balances. To obtain a flux three measurements are required: (a) compound concentration in sediment on a dry weight basis, (b) sedimentation rate, and (c) in-situ density of sediment. Previous work indicates that a typical flux for polycyclic aromatic hydrocarbons in Lake Superior is about 0.3 ng cm^{-2} yr^{-1}. All of these measurements will be made on sediment core samples sliced at centimeter intervals. Extracts will be fractionated on silica gel and concentrations of compounds determined by selected ion monitoring mass spectrometry. Sedimentation rates will be determined using lead-210, cesium-137 and ragweed pollen. In-situ density measurements will be made using standard procedures. Measurements will be obtained on 40 core samples taken at different locations to allow a mapping of fluxes in the Lake for each compound. Fish and benthic organisms will also be studied to provide a picture of bioaccumulation.

Expected Results of Research:

The basic information to be obtained is a contour mapping of the fluxes to the sediment of about 20 organics from the dumpsites and bioaccumulation in fish and benthic organisms. Because of the unique nature of some of these compounds the movement from dumpsite to sediment can be traced. Rough mass balances will be obtained from the flux measurements to provide an approximation of the amount of material from the dumpsites which has entered the lake sediments. Data from samples taken in later years will allow disappearance of these compounds from the system to be determined.

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Title:
Developments at the Eleventh International Conference on Water Pollution Research

Principal Investigator:
F. G. Pohland

Institution:
United States of America National Committee for the International Association on Water Pollution Research

Project Period:
1 year

Budget:
(1 year) $52,260

EPA Project Number:
R 809703

This project provides a report which gives a current assessment of developments in water pollution research throughout the world. The report is based on information gathered at the Eleventh International Conference on Water Pollution Research, held at Cape Town, South Africa, March 29 - April 2, 1982. The Conference brought together research engineers and scientists, consulting engineers, manufacturers of process control equipment, administrators of water quality control programs and others involved in water pollution control. Approximately 30 of the engineers and scientists attending the Conference from the USA participated in the preparation of this report. Each participant submitted a summary on developments at an assigned session of the Conference for inclusion in the overall report. Both formal and informal developments are covered, including pertinent comments and ideas brought out in discussion of presented papers and among attending researchers. Copies of the report, "Development at Eleventh International Conference on Water Pollution Research," dated February 1983 are available from the EPA.

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Title:
The Guidance of Airflow by an Isolated Valley

Principal Investigator:
Ronald Dobosy

Institution:
Iowa State University

Project Period:
1 year

Budget:
(2 years) $49,567

EPA Project Number:
R 809588

Several studies in moderate-relief complex terrain have found that valleys guide air flow. Parameterization of this guidance should improve plume trajectory forecasts in dispersion models. To be applicable in general terrain the parameterization should be derived from basic physical understanding; this must begin from careful study of a simple valley isolated from other terrain.

We have evolved a hypothetical explanation of the valley effect which we propose to test and refine, starting with a numerical model adapted from existing models. The numerical results will be used to develop a detailed field plan to verify and further refine the hypothesis. We intend to execute the field plan in a subsequent project, but limited initial tests with existing equipment are planned for this project.

The field plan will apply to a straight section of the Des Moines River Valley in central Iowa, a 75-m deep 1000-m wide valley in terrain otherwise flat except for widely separated creeks. The relative simplicity of this terrain will allow our findings and plans to be useful to similar projects.

Title:
Theory and Applications of Direct Filtration

Principal Investigator:
Charles R. O'Melia

Institution:
Johns Hopkins
University

Project Period:
3 years

Budget:
(2 years) $104,323

EPA Project Number:
R 810094

The general objective of this research is to relate the design of direct filtration facilities in water treatment to the quality of the raw water to be treated. More specifically, interrelationships among packed bed filter performance as characterized by effluent quality and head loss development, flocculation facilities as described by velocity gradients and reaction time, and raw water quality as represented by solids concentration and particle size distribution will be assessed, developed as needed, evaluated theoretically and experimentally, and appraised as bases for design.

The work is divided into two phases. The first phase involves adaptation of existing concepts to the direct filtration process and the use of sensitivity analyses to identify important characteristics of the process. The second phase entails experimental testing and application of the model and the development of bases for process design.

The expected results have potential for providing improved methods for the design and operation of direct filtration facilities.

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Title:
Aerosol Deposition in the Human Oral Passage

Principal Investigator:
David L. Swift, Ph.D.

Institution:
Johns Hopkins
University School of
Hygiene and
Public Health

Project Period:
2 years

Budget:
(2 years) $155,914

EPA Project Number:
R 809074

The overall objective of this research program is to determine the percent deposition of inhaled aerosols in the human oral passage under normal breathing conditions as a function of the breathing rate, aerosol particle size, and oral passage configuration. The objective is to be met by a series of experimental studies in normal human subjects in which radiolabelled monodisperse particles will be inhaled at controlled conditions by mouth without a breathing tube. This period of inhalation will be followed by a measurement of deposition employing a gamma scintillation camera. The results of the study will be a series of curves of percent oral passage deposition vs. particle size depending on the oral passage configurations and/or airway resistance.

The project has been underway since September 1982; equipment is in construction but no experiments have been carried out at present.

Title:

Evaluation on the Toxicologic Effects of Environmental Carcinogens with Human Tissue Model Systems

Principal Investigator:
Ih-Chang Hsu

Institution:
Department of Pathology
University of Maryland School of Medicine

Project Period:
2 years

Budget:
(2 years) $148,089

EPA Project Number:
R 809835

The objectives of this proposed research are to determine the genotoxic hazards of the aromatic nitrogen pollutants, i.e., naphthylamines, aminoanthracenes, aniline, aminobiphenyls, aminopyrenes and nitropyrenes, on target cells or tissues of different animal species including humans. Metabolism, activation and genotoxic effects of these carcinogens in these tissues or cells will be compared so as to serve as a valid basis for extrapolation from animal data to humans.

Human pulmonary alveolar macrophases (PAM), bronchial explants, and hepatocytes of human and rats are now routinely prepared in my laboratory. Some of the cells are being treated with aminopyrene, aminoanthracene, naphthylamine, aminobiphenyls, etc. The active metabolites generated by the cells will be measured by the mutation frequency in co-cultivated indicator Chinese hamster V79 cells. Unscheduled DNA synthesis and carcinogen DNA binding will also be determined in the treated cells for comparing DNA damage. The metabolites in the media will also be quantitated and evaluated for the relationship to their genotoxic effects. Since determination of metabolites and carcinogen DNA adducts are very time consuming, especially without radiolabeled compounds, it is unlikely to determine the adduct and metabolites in 2 years for all aromatic amines listed above. We will focus on aminopyrene and nitropyrene and label them with tritium for comparing adduct formation and metabolism in the cells of humans and rodents. Nitropyrene has been demonstrated recently to be the most important genotoxic compound in diesel fuel emissions. It is too early to predict the difference of genotoxicity in cells of different animal species. Parallel experiments for measuring these effects are being conducted in human and rodent cells so that the comparison of the results can provide a link between animal cancer studies and human cancer.

Title:
Interspecies Comparison of Liver Carcinogenesis

Principal Investigator:
Dr. Michael Lipsky

Institution:
University of
Maryland
Baltimore, MD

Project Period:
3 years

Budget:
(2 years) $259,967

EPA Project Number:
R 809599

Objectives:

The overall goals of the proposed research are: (A) To investigate quantitative relationships between molecular and subcellular events presumed to be important in the carcinogenic process (such as the amount of carcinogen-DNA adducts formed) and end points of organismal response (such as the number of neoplasms formed) in a target organ; (B) To develop a scientifically sound basis for reliable, quantitative extrapolation of data from animal studies to man. This will be accomplished by comparing unscheduled DNA synthesis (UDS), metabolism, carcinogen-DNA adducts, mutagenic activity, preneoplastic lesion formation and neoplasm formation between different species including rats, mice and rainbow trout *in vivo* and between primary hepatocyte culture from humans, rats, mice and trout *in vitro* using aflatoxin B1 and diethylnitrosamine which are positive hepatocarcinogens in some species and negative in at least one.

Expected Results of Research:

The proposed research will provide new insights into quantitative relationships between different events presumed to be important in the process of chemical carcinogenesis. This project will add new data to help validate or question the validity of the somatic mutation hypothesis of chemical carcinogenesis. In terms of the general populations, the proposed research will help establish a valid scientific basis for quantitative risk assessment and extrapolation from animal data to man. Although a great deal of federal regulation and legislation is directly related to risk assessment of environmental hazards and quantitative extrapolation from animal studies to the human situation, little hard scientific data is available to lend support and guidance to the extrapolation process. The proposed research will be a step to establishing this scientific base. This can have implications in the legislative and regulatory processes, and in evaluating human risk to provide a basis for primary cancer prevention.

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Title:
Insufficient Disinfection of Water Supplies Attributable to Chlorine Residual Monitoring

Principal Investigator:
Neil M. Ram, PhD

Institution:
University of Massachusetts/ Amherst

Project Period:
2 years

Budget:
(2 years) $60,209

EPA Project Number:
R 809767

A secure hygienic quality in water supplies is dependent upon the maintenance of a free chlorine residual. The available analytical methods used to differentiate between free and combined chlorine, however, are subject to interference from nitrogenous organic compounds. An apparent residual measured as 0.2 mg/L "free chlorine" may therefore not provide a sufficient level of disinfection in the presence of N-organic compounds. While it is possible to calculate the contribution of these N-organic materials to falsely positive tests for free chlorine, this determination requires prior knowledge of the exact concentration and types of N-organic compounds present as well as their relative specific rates of formation. The objective of this study, therefore, is to determine the true level of disinfection, as measured by the percent kill of *Escherechia coli*, in water containing N-organic compounds exposed to supposedly adequate levels of disinfectant measured by the DPD analytical determination for free chlorine. The decreased kill of *E. coli* attributable to the presence of nongermicidal chloroorganic compounds yielding falsely positive values of free chlorine, would provide data to determine if sufficient disinfection of water supplies is achieved by free chlorine residual monitoring. While previous studies have demonstrated the inability of existing analytical methods to differentiate between free and N-chloroorganic compounds and the nongermicidal behavior of these N-chloroorganic substances, no investigation has actually examined the possibility of inadequate disinfection of water supplies containing such N-organic precursors attributable to free chlorine residual monitoring. Since suppliers of water may substitute the use of chlorine residual monitoring (equal to 0.2 mg/L) for up to 75 percent of the bacteriological samples, information about the actual level of disinfection attributable to possible false positive free chlorine values, is needed to insure a hygienically safe water. Furthermore, total organic nitrogen will be examined as a possible surrogate parameter for the level of interference of N-chloroorganic substances in disinfection.

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Title:
Microbial Transformation Rates and Fate of Polycyclic Aromatic Hydrocarbons in Estuarine Sediments

Principal Investigator:
Michael P. Shiaris

Institution:
University of Massachusetts Boston Harbor Campus

Project Period:
2 years

Budget:
(2 years) $189,508

EPA Project Number:
R 810119

The ongoing investigation will demonstrate the interaction of microbiological and physico-chemical processes in the transformation and fate of polycyclic aromatic hydrocarbons (PAH) under natural field conditions. Experiments will be undertaken to determine the *rates*, the *mechanism(s)*, the *microbial ecology*, and *effects* of *varying environmental factors affecting PAH transformation* in *estuarine and coastal sediments*. Radiotracer and microbiological techniques, gas-liquid chromatography, thin layer chromatography, gas chromatography/mass spectrometry and high pressure liquid chromatography will be employed to analyze and identify potential PAH metabolite accumulation. Enumeration and identification of predominant PAH-degrading microorganisms, their ability to transform a variety of PAH compounds, and their potential rates of PAH degradation will be compared to *field* and *simulated-sediment experiments*. Biochemical probes will be employed to determine the major microbial communities involved in PAH decomposition and the pathways of PAH degradation that may occur *in situ*. The major objectives and expected results of this investigation are to determine the concentration of PAH in key urban and pristine coastal sediments; to estimate the specific biodegradation rates in sediments; to identify the major environmental factors affecting biodegradation; to assess the fate of model PAHs in coastal sediments; and to enumerate and identify the predominant microorganisms involved in sediment PAH transformation and evaluate their ability to degrade a variety of aromatic compounds.

Emphasis is placed on the integration of field and laboratory PAH biodegradation experiments and relevant microbial ecology of this dynamic process. Accomplishment of these objectives will aid in the understanding of microbial transformation of PAHs and related aromatic compounds in coastal sediments.

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Title:

Identification and Quantification of the Principal Transport Mechanisms Responsible for Movement of Toxic Organics in Saturated Groundwater Flow

Principal Investigator:
John C. Crittenden
and Neil J. Hutzler

Institution:
Michigan Technological University

Project Period:
2 years

Budget:
(2 years) $172,100

EPA Project Number:
R 809464

Describing the transport of toxic organics in the soil environment is essential for proper site selection for land application and disposal sites for hazardous wastes and assessing groundwater contamination resulting from accidental spills. The work proposed herein will examine the mechanisms that affect the transport and attenuation of toxic organics in saturated groundwater flow. A mathematical model which incorporates the following rate nd mass transfer mechanisms will be used to evaluate the importance of each mechanism: (a) biodegradation, (b) adsorption, (c) dispersion, (d) channeling, and (e) facilitated transport by organic complexation with humic substances. Column data derived with a number of soil matrices, organic chemicals, and aqueous-phase solution conditions will be compared to model predictions and the ability of the model to predict quantitatively the transport of toxic organics will be tested. A methodology for determining model parameters from batch rate, equilibrium, and other tests will be developed and these tests will also be used to examine and formulate the mathematic descriptions of each mechanism. Once the one-dimensional transport of toxic organics are well described in this study, the mathematical relationships which were used to describe the mechanisms and the methodology which was used to determine model parameters can be used in two- and three-dimensional flow problems as would be encountered in the field.

Title:
Measurement of HO_2 (RO_2) in Ambient Air

Principal Investigator:
Donald Stedman

Institution:
University of Michigan

Project Period:
2 Years

Budget:
(2 years) $159,483

EPA Project Number:
R 808706

It has been shown that HO_2 (RO_2) detection is possible using a chemical amplifier based on the scheme:

$$HO_2 + NO \rightarrow OH + NO_2$$
$$OH + CO \rightarrow H + CO_2$$
$$H + O_2 + M \rightarrow HO_2 + M$$

and that an appropriate species for the detection of HO_2 (RO_2) is the NO_2 formed in the chain reaction enhanced by the addition of controlled amounts of CO and NO to ambient air. The best detector of NO_2 is a chemiluminescence detector based on reaction with luminol. This detector system has been demonstrated for HO_2 ambient air, but needs optimization, testing, and controlled field studies.

Calibration of the system will be carried out using several smog chamber facilities and by means of chlorine photosensitized HO_2 formation. The objective of this program is to produce the first direct continuous measurement of HO_2 (RO_2), species which are fundamental to an understanding of air pollution chemistry.

Title:

Parameter Evaluation for Predictive Modeling and Design of Multicomponent Systems

Principal Investigator:
Walter J. Weber, Jr.

Institution:
University of
Michigan

Project Period:
3 years

Budget:
(2 years) $207,796

EPA Project Number:
R 809808

The design of fixed-bed adsorbers utilizing activated carbon or synthetic resins for removal of organic contaminants in water and waste treatment requires estimation of when an effluent will reach a specified breakthrough stage or concentration level for a specific set of influent conditions. Predictive models which describe adsorber effluent concentration profiles can be extremely valuable in this regard, as well as being useful as the interpretive elements of feed-forward and feed-back control systems for adsorber operation.

Waters and wastewaters generally contain a number of adsorbing organic species; that is, they are multicomponent systems. Reliable determination of the corresponding series of equilibrium and mass transport parameters required as input for predictive modeling of the behavior of multiple components in fixed-bed adsorbers is at best difficult, and at times impossible. Two methods that represent potentially major breakthroughs in this field have surfaced recently for single-component systems. The high pressure mini-column (HPMC) technique and the micro-column technique provide increased simplicity and greater accuracy in the determination of equilibrium and mass transfer coefficients, respectively, for adsorber modeling.

The proposed research will extend these methods to the more common and significant situation of multicomponent adsorption. Principal effort will be focused on the development of experimental methods that can evaluate the interactive vis-a-vis independent components of equilibrium and mass transfer parameters. This effort is inextricably linked to the development of appropriate mathematical relationships describing multiple solute interactions. The expected results of the work will be development of a procedure which couples experimental methodologies with an interpretive mathematical model to facilitate the design and operation of adsorption systems for water and waste treatment.

Title:
Kinetics of Oxidation of Large Organic Compounds Relevant to the Incineration of Toxic Wastes

Principal Investigator:
Frank T. Greene

Institution:
Midwest Research Institute

Project Period:
2 years

Budget:
(2 years) $444,304

EPA Project Number:
R 808184

The objective of this research is the determination of the rates and mechanisms of oxidation of organic compounds of the types of importance in toxic wastes. The reactions will be carried out in high temperature flow reactors and in laminar flat flames burned at both reduced and atmospheric pressure. These systems are complementary in that the flow reactors will provide particularly well-defined isothermal conditions while the flat flames provide more typical combustion conditions. The reaction products and intermediates, including free radicals and other unstable and reactive species, will be identified and measured as a function of time principally by molecular beam mass spectrometry. In addition, optical spectrometry, microprobe, and other techniques will be used as needed to obtain supplementary data. Temperatures will be measured by molecular beam, thermocouple, and spectroscopic techniques. Flow fields will be characterized by conventional and time-resolved schlieren photography. This ensemble of measurements is expected to provide more detailed kinetic data than has been previously available for the complex molecules. These data will be evaluated to determine optimum incineration conditions and possible alternative oxidation processes.

Title:
The Role of Chloramines in the Oxidation of Organic Compounds Present in Water During Disinfection

Principal Investigator:
Robert M. Carlson

Institution:
University of
Minnesota
Duluth, MN

Project Period:
2 years

Budget:
(2 years) $154,921

EPA Project Number:
R 809695

The proposed project will involve the determination in aqueous media of the product distribution and, where appropriate, the kinetics for the reaction of inorganic (NH_2Cl, $NHCl_2$, HCl_3) and organic chloramines with representative examples of several classes of environmentally significant organic compounds. These classes will include polynuclear aromatic hydrocarbons (PAH) and their heterocyclic analogs, phenols, aldehydes, ketones, amides, and aromatic amines. For each compound where rate data is obtained, the results will be described by equations which will allow the prediction of the rates of reaction of the compound under conditions encountered during disinfection processes. The results obtained in the processed study for PAH compounds can be directly compared to the results gathered in two related ongoing projects in our laboratory involving the aqueous reactions of PAH with chlorine and chlorine dioxide.

The effect of various parameters that may substantially affect the type and distribution of organic products generated during aqueous chloramine reactions will also be addressed. These will include such parameters as the effect of pH, dissolved oxygen, and the presence of oxidative chlorine species (e.g., ^-OCl, $HOCl$, Cl_2, ClO_2, ClO_2^- and ClO_3^-). The investigators will also be vigilant for factors such as micelle formation, the presence of particulate matter, or light-initiated reactions that in earlier investigations were observed to confound the interpretation of the results.

The products identified in the proposed investigation will be made available (by purchase or synthesis) for biological testing under a potential cooperative arrangement with the Health Effects Research Laboratory, U.S. Environmental Protection Agency, Cincinnati, Ohio. Moreover, these pure compounds will represent standards for quantitation in the proposed research as well as for the analysis of environmental samples by gas chromatography/mass spectrometry (GC/MS).

The project will also examine reactions that closely match actual field conditions. For example, a comparison will be made of the product distributions in the reactions of oxidative chlorine species and PAH observed in the laboratory with samples of chlorinated water obtained from tar-coated pipes common to many municipal distribution systems. The possible evaluation of other situations where chloramines may interact with organic chemical additives (e.g., coagulants) will be evaluated as the project develops.

Title:
Acute Toxicity of Fossil Fuel Hydrocarbons to Lemna

Principal Investigator:
Joe M. King

Institution:
Murray State University

Project Period:
2 years

Budget:
(2 years) $61,682

EPA Project Number:
R 809946

This project is designed to determine the acute toxicity of water soluble fractions of select crude oils, refined oils, coal-liquefaction products, and shale oil to species of *Lemna (L. gibba, L. minor, L. perpusilla)*. Effects of photoperiod, pH, and temperature on the sensitivity of these organisms to fresh and aged extracts will be determined. Pure components of the water soluble fractions will be tested to identify those which are most toxic to *Lemna*.

Tests will be conducted with axenic cultures of *Lemna* in a chemically defined growth medium to which various levels of extracts or pure compounds are added. Inoculated test solutions will be incubated in controlled environment chambers set at the desired light intensity, photoperiod, and temperature. All tests will be run in triplicate with controls. The number of fronds in each culture will be determined daily for 14 days and growth rates will be measured as the slope of the straight-line portion of the semi-log plot of frond numbers versus time. A toxicity curve will be constructed by plotting toxicant concentration versus reduction in growth rate as a percent of the controls.

This study will provide insight into the use of *Lemna* as a bioassay organism for aquatic toxicants, such as petroleum hydrocarbons, which accumulate at the air-water interface. Data obtained will indicate the relative sensitivities of the test organisms to petroleum hydrocarbons under various environmental conditions.

Title:

Methylated Metal and Metalloid Compounds and Water Quality

Principal Investigator:
James H. Weber

Institution:
University of
New Hampshire

Project Period:
3 years

Budget:
(3 years) $56,602

EPA Project Number:
R 809416

The objectives of this research are to study the presence, formation, and stability of methylated metal ions in natural waters so we can better assess their concentration and movement in systems such as domestic water supplies and waste water. There are several types of experiments that will aid us in achieving the objectives. In all experiments we will use techniques such as gas chromatography, atomic emission spectroscopy, and/or atomic absorption spectroscopy. (1) We will determine concentrations of methylated metal ions in natural waters. (2) We will measure the rates of decomposition of methylated metal ions and the rates of formation of products from the laboratory. (3) We will attempt to learn the mode of formation of methylated metal ions in a variety of natural water samples.

Title:

Thermal and Catalytic Reactions of Hydrocarbons with Water Vapor and Hydrogen

Principal Investigator:
Dr. Joseph W. Bozzelli

Institution:
New Jersey
Institute of
Technology
Newark, NJ

Project Period:
3 years

Budget:
(2 years) $143,300

EPA Project Number:
R 810014

This research project will investigate chemical reaction kinetics on detoxification reactions of chlorinated hydrocarbons such as carbon tetrachloride, PCBs, DDT, other pesticides, and herbicides using water vapor (steam) as a reagent. The halocarbons will be converted through this reaction scheme into low BTU fuel gas (Hydrocarbons and Carbon monoxide) plus HCl, which can be readily neutralized. The conversions will be examined in two different types of chemical reactors:

1. A high temperature tabular flow reactor to study the reactions under homogenous vapor phase conditions.
2. A medium temperature catalytic reactor to effect the above detoxification and possibly the in-situ neutralization of HCl product.

Previous research on organic halocarbon reactions has dealt with oxidation, which results in production of unstable chlorine compounds. This project will introduce hydrogen in the form of water vapor as a reactant to obtain HCl, a stable, easily neutralized end product.

The main objectives in this study are:

1. To determine the overall kinetics of chlorinated hydrocarbons' reactions with water vapor and hydrogen;
2. To develop a thorough understanding and knowledge of these reactions, the initial products, subsequent reactions and most importantly, the final products.

The reactions will be studied in temperature controlled tubular or catalytic reactors with complete end product and reagent analysis by on-line flame ionization gas chromatography (FID 1C), GC/Mass Spectrometry and carbon monoxide analyses. Kinetic parameters will be determined as a function of reagent residence time, temperature and reactant concentrations.

The results from the high temperature tubular flow reactor are applicable to currently operating incinerators where a stream input would effect complete conversion of chlorinated species to HCl; with energy for steam generation resulting from heat exchange on the incinerator. Presently available coal gasification reactors and pilot plants could be directly utilized for these homogenous reactions without significant changes.

Results from the heterogenous catalytic reaction studies could lead to efficient detoxification of halocarbons at lower temperature and may provide a useful technique for decontaminating leachates and/or polluted ground waters where these streams would be heated (not vaporized) and run through a catalytic reactor.

Title:
Interactions Between Microbiota and Persistent Pollutants in Marine Sediment

Principal Investigator:
Douglas G. Capone

Institution:
State University of
New York

Project Period:
2 years

Budget:
(2 years) $92,055

EPA Project Number:
R 809475

The upper strata of marine sediments are the locus of enhanced microbial biomass and activity. Microorganisms which participate in aspects of several (e.g. carbon, nitrogen, sulfur) important material cycles reside in these environments. Marine sediments are also the site of an ever increasing load of persistent pollutants, such as chlorinated hydrocarbons and heavy metals. We are presently investigating the effect of several common contaminants on microbial viability (measured as ATP biomass and epifluorescent counts) and selected microbial activities in unimpacted estuarine and salt marsh sediments. Bacterial methanogenesis, sulfate reduction, N_2 fixation, denitrification, thymidine uptake, and heterotrophic potentials are being tested for sensitivity to these compounds.

We herein propose to extend these studies to include comparisons of the microbial biomass and predominant respiratory pathways of relatively pristine through heavily impacted sediments, as a function of the pollutant loading. Further, we intend to determine the comparative rates and pathways of mineralization of selected organic pollutants in differentially impacted sediments. Finally, we propose to investigate the interrelationships of the meiofaunal and bacterial populations of these sediments, with respect to the capacity of the bacterial flora to mineralize specific compounds.

The results of these studies should help determine the consequences of pollutant loading to the microbiota of marine sediments. Sensitive pathways will be identified and evaluated for their value as diagnostics of the status of a particular sediment or for possible bioassay application. Lastly, the studies of organic pollutant mineralization will provide insight into the capacity and potential mechanisms of detoxification by these sediments.

Title:
Pathology of Small Mammals: Index of Toxicity at Waste Dumps

Principal Investigator:
Dr. John J. Christian

Institution:
SUNY Binghamton

Project Period:
2 years

Budget:
(2 years) $175,540

EPA Project Number:
R 809437

Our objective is to determine if small indigenous mammals may serve as indicators of environmental pollution and potential health risks at chemical waste dumps, and to monitor such dumps. Small mammals, mainly voles (*Microtus pennsylvanicus*), will be live-trapped at Love Canal, its immediate surroundings, and at uncontaminated "reference"areas. Body and liver weights will be obtained. Samples of the liver will be prepared for EM, using standard procedures. Liver will be frozen for measurement of hepatic AHH, LDH, SGOT, SPGT, and glycogen. Reproductive data, size of embryo, number of embryos, resorbing embryos, uterine placental scars and number of corpora lutea, state of the uterus, and position of the testes will be recorded. Organic pollutants in fat will be determined by HPLC-MC by S.R.C. Other organs will be preserved in 4% paraformaldehyde. Weights of reproductive organs, adrenals, thymus, and spleens will be obtained after fixation. Pieces of the various organs will be prepared for light microscopy. Procedures to be used have been defined in preliminary studies. Dry weight of the eye lenses will be obtained for determining relative age.

Demographic data such as relative densities, reproductive rates, age composition, etc. will be compared between areas and with data available from uncontaminated areas. We have developed these procedures and have preliminary baseline values.

Pens containing laboratory raised voles will be placed in presumptive contaminated and reference areas, and the voles removed after various lengths of time. The same procedures will be used to evaluate these animals.

Once a data base is obtained for one dump, we will repeat the procedures at other selected dumps.

We expect that the demographic data will reveal changes in reproductive parameters, mortality rates, density and age-composition of populations from polluted areas with respect to reference areas. Voles from areas close to might be expected to indicate whether pollutants have migrated from the dump and should show changes between those from the dump-site and reference areas.

Identification pollutants in fats and their relative amounts should provide indices of contamination of the animals and should relate to pathological changes in their organs. These data should provide a useful way of evaluating potential health hazards. The route of contamination of the animals would be through contact and by ingestion of contaminated vegetation, invertebrates, and ground water.

Title:

Chemical Degradation of Carbamate Pesticides: Water Detoxification Using Reactive Ion Exchange

Principal Investigator:
Gilbert E. Janauer

Institution:
SUNY-Binghamton

Project Period:
2 years

Budget:
(2 years) $142,307

EPA Project Number:
R 809867

It is the primary objective of this research to demonstrate the feasibility of a new, safe detoxification method for carbamate pesticides. Since carbamates such as Aldicarb, Methomyl, Oxamyl, and Carbofuran are very effective and nonpersistent crop protectants, their continued and future use is anticipated. It has been found, however, that special circumstances or accidents may result in the inadvertent intrusion of such compounds into (ground) water supplies intended for human consumption at levels higher than accepted by current guidelines (e.g., in NY state). *In situ* detoxification at the point-of-use is to be accomplished by acid or base hydrolysis (which may follow prior oxidation) during mere passage (of the contaminated water) through a column or cartridge packed with reactant ion exchange resinates or other insoluble, high capacity reactant-carrying material.

Several major subordinate objectives must be met in order to reach the primary goal. This will be accomplishd by:

1. Acquisition of information on identity and ultimate fate of degradation products of Aldicarb and other pesticides during nucleophilic cleavage/hydrolysis and oxidation in aqueous solutions and on reactive ion exchange resinate columns.
2. Determination of hydrolysis and oxidation rates for candidate pesticdes in solution and on resin beds under realistic experimental conditions (pH, T, water hardness, bed contact time → flow rate in columns, etc.).
3. Adaptation and improvement of current methodology for quantitation and monitoring of Aldicarb and other carbamates, and their reaction products. Good precision and quantitative yield at the nanogram level are a prerequisite to the success of this work.

Methods used in reactive ion exchange experiments encompass straightforward batch contact and flow-through (column) procedures with emphasis on careful monitoring of parent pesticides and all degradation products to achieve a complete mass balance on column influents vs. effluents. For quantitation at the ppb level the use of preconcentration/HPLC, GC-MS in SIMS mode, and, possibly, HPLC-MS is anticipated. Initial results indicate that Aldicarb, Carbofuran and their metabolites can currently be detected at this level using GC with a thermionic detector. Base and acid hydrolysis rates in solution are being determined using this analytical methodology, while HPLC is being used to acquire data in conjunction with parallel reactive ion exchange experiments under identical or similar conditions.

The rate of hydrolysis results for candidate pesticides in solution will be correlated with the corresponding reactive ion exchange rate data (pseudo first order rate constants) obtained in batch and columns under reproducible and representative conditions. This will permit, eventually, fairly accurate predictions of column detoxification as a function of system variables.

Finally, quantitative results on *in situ* detoxification efficacy in terms of rate of degradation (and removal by sorption of decomposition products) will be available. These will, hopefully, permit the Principal Investigator to design a preprototype assembly for a practical carbamate pesticide detoxification unit.

The results of the entire study will also be of potential benefit in cleanup of accidental spills (pesticide transportation), safe disposal, and protection of workers handling carbamates. Furthermore, there may be progress made with respect to analytical methodology, and the solution studies may shed light on mechanisms of carbamate degradation in the environment. Last, but not least, it should be mentioned that many organophosphorus pesticides can be detoxified using exactly the same approach.

Title:
Reactions of Adsorbed Polycyclic Aromatic Hydrocarbons

Principal Investigator:
Joan Darsey

Institution:
New York University
Medical Center

Project Period:
3 years

Budget:
(2 years) $282,454

EPA Project Number:
R 810099

This is an investigation of the reactions of polycyclic aromatic hydrocarbons (PAH) adsorbed on particles, under simulated atmospheric conditions. Fourier-transform Infrared Spectroscopy (FTIR) (transmission and photoacoustic) will be used to characterize the particle surfaces and to study the interactions of adsorbed PAH with the surface. The objectives of the study are: 1) to characterize adsorbed PAH and their degradation products, *in situ* by FTIR spectroscopy and by extraction and high pressure liquid chromatography (HPLC); 2) to determine the rates of degradation of PAHs adsorbed on selected particle substrates under simulated atmosphere conditions, using the fluidized-bed photoreactor developed for such studies; 3) to identify the products of degradation of benzo(a) pyrene on each substrate (by HPLC) to gain insight into the possible mechanisms of reaction; and 4) to identify key variables which influence the rates and products of PAH degradation. The substrates to be investigated include coal fly ash, carbon, silica, alumina and hematite. Reactions will be studied in air, in the presence and absence of visible light, and with added gases. The effects of temperature and PAH concentration on the surface will be examined.

Increased coal and wood-burning in the U.S. can result in greater emissions of PAH. The proposed study will provide a more fundamental understanding of the critical factors which influence the chemical and biological properties of airborne PAH and their lifetimes in the atmosphere. The study will also provide improved techniques for *in situ* studies of adsorbed PAH.

Title:

Cocarcinogenicity of Inhaled Sulfur Dioxide and Endogenous Sulfite with Benzo(a)pyrene

Principal Investigator:
Albert F. Gunnison

Institution:
New York University
Medical Center

Project Period:
3 years

Budget:
(2 years) $250,000

EPA Project Number:
R 809860

The major objectives of this project are to compare the effect of 1) inhaled sulfur dioxide (SO_2) and 2) endogenously-generated sulfite/bisulfite (the hydrate of SO_2 at physiological pH) on the time of appearance and incidence of squamous cell lung cancers induced in rats by exposure to benzo(a)pyrene (BaP). The studies should throw light on possible mechanisms of action of SO_2/sulfite in the carcinogenic process.

Young adult male Sprague Dawley rats will be exposed for 15 consecutive weeks to BaP by intratracheal instillation at a dose of 1 mg/rat/week. Some of the rats will be exposed by inhalation to 10 and others to 30 ppm SO_2 for a period of 21 consecutive weeks, beginning 2 weeks prior to the intratracheal instillations. Additional experimental groups will be exposed to SO_2 alone and one group instilled with the vehicle alone will serve as a control. A parallel experiment using sulfite oxidase-deficient rats will be conducted simultaneously. In these animals sulfite will be present systemically due to endogenous generation. Tracheal sulfite and S-sulfonate ($RS\text{-}SO_3^-$) concentrations will be determined in all experimental groups and used as a basis for establishing sulfite dose to the trachea. Animals will be allowed to live out their natural lives and, at the time of death, slides will be prepared of the lungs and of all tumors.

Based on data in the literature, the proposed exposure of rats to BaP is expected to yield an incidence of squamous cell lung cancer of approximately 20%. Groups which are simultaneously exposed to SO_2 or to endogenously-generated sulfite should have either the same or a higher incidence and/or shorter induction period depending upon the degree of augmentation by sulfite. Animals which are exposed only to inhaled SO_2 or endogenously-generated sulfite are not expected to develop lung cancers. Among the BaP-instilled groups, any differences in the number or time of appearance of tumors which correlate with the mode of sulfite exposure should provide information on its mechanism of action.

Title:
Effects of Inhaled Sulfuric Acid on Healthy Humans

Principal Investigator:
Morton Lippmann, PhD

Institution:
New York University, Institute of Environmental Medicine

Project Period:
2 years

Budget:
(2 years) $222,326

EPA Project Number:
R 810101

In a previous study at this laboratory, the bronchial mucociliary particle clearance of healthy nonsmokers was markedly altered after a one-hour inhalation of a submicrometer sized sulfuric acid (H_2SO_4) aerosol at a concentration of 100 $\mu g/m^3$. We found no measurable changes in respiratory mechanical function for exposures up to 1,000 $\mu g/m^3$, demonstrating that clearance measurements provided a more sensitive tool to determine physiological effects of such exposures.

The nature of the transient effect on clearance which was observed, and the locus of response in relation to deposited doses in the airways, will be determined in further tests on human volunteers exposed for one- and two-hour periods to 100 $\mu g/m^3$ of H_2SO_4. The subjects will be exposed to γ-tagged monodisperse Fe_2O_3 aerosols both before and after the acid aerosol exposure, and the retention of both tagged aerosols will be measured periodically with external detectors. By comparison of the retention times in the acid exposure tests with those measured in sham exposure and control tests, we will determine the effects of the acid exposure on mucus transport velocities at each airway level and on the rate of clearance from the alveolar region of the lungs.

Title:
Dephenolization of High Strength Phenolic Wastewaters by a Catalyzed Aerobic Coupling Reaction

Principal Investigator:
P. K. Lim

Institution:
North Carolina
State University

Project Period:
2 years

Budget:
(2 years) $173,164

EPA Project Number:
R 809553

The primary objective of the proposed study is to verify the viability of a novel dephenolization scheme which is based on a catalyzed, aerobic coupling reaction. The proposed scheme is designed to treat high strength phenolic liquors which are generated in coal-conversion processes.

As envisioned in the proposed scheme, phenols will be converted catalytically and aerobically into insoluble coupling products which can then be easily separated from the wastewaters. The attractive features of the scheme include: (i) the ability to treat concentrated phenolic wastewaters without dilution, (ii) low costs of oxygen and catalyst, (iii) simplicity of the scheme, (iv) easy disposal of recovered solid organics as a fuel.

Various transition metal compounds and complexes, including cuprous chloride, will be screened for their ability to effect the desired aerobic coupling reaction under mild reaction conditions. Preliminary studies have shown cuprous chloride to be a potent coupling catalyst. The kinetics of the catalyzed coupling reaction will be studied, and the major coupling products will be characterized and identified. The effects of potential inhibitors will be examined and optimum reaction conditions for the coupling reaction will be sought and then verified with actual coal gasification wastewaters. Finally, a cost analysis of the novel dephenolization scheme will be carried out.

Title:
Ultraviolet Disinfection of Water and Wastewater

Principal Investigator:
Dr. J. Donald Johnson

Institution:
University of North Carolina
Chapel Hill

Project Period:
1 year

Budget:
(1 year) $72,012

EPA Project Number:
R 809593

Objectives:

Ultraviolet light is being investigated as a water and wastewater disinfectant which produces none of the toxic by-products typical of chlorine disinfection. It is the purpose of this study to define the UV energy required to inactivate microorganisms of human health interest likely to be present in water and wastewater. The rates of kill of these organisms will be measured for *E. coli, Streptococcus faecalis, Staphylococcus aureus, Salmonella typhi, Shigella sonnei, B. subtilis, rotavirus, poliovirus,* and *entamoeba moshkovskii,* and natural coliforms. Comparisons will be made of the relative sensitivities of these organisms to the natural water indicator organism, *E. coli,* as an indicator of the resistance of these organisms to ultraviolet disinfection in comparison to the relative resistance they have to coliform disinfection with chlorine. The response of these organisms to ultraviolet radiation dose will be measured as the log survivals as a function of water quality, especially in municipal wastewater effluents undergoing various treatments. The importance of filtration prior to ultraviolet disinfection and of turbidity on ultraviolet disinfection will be investigated by measuring the effects of particulate size and size fractions on particle size distribution after and before filtration, as well as the ultraviolet dose-survival curves in typical municipal wastewater.

Expected Results of Research:

This study will accurately determine the quantity of ultraviolet radiation required for inactivation of a range of organisms of public health importance. This will provide the necessary information to evaluate ultraviolet light as an alternative to chlorination for the inactivation of these organisms in natural water systems. The accurate determination of the quantity of ultraviolet radiation required to produce inactivation of waterborne, infectious organisms will provide the basic data necessary to design efficient ultraviolet disinfection units for wastewater and drinking water disinfection. Data on the effects of turbidity and particulates in water disinfection will determine the necessary water quality for effective ultraviolet disinfection. Since particulate protection can be a major factor in limiting disinfection, the quantitative data on particle size distribution of particulate material and its effect on the rates of ultraviolet inactivation of microorganisms will be used to evaluate what degree of pretreatment or filtration is required for effective ultraviolet disinfection.

Title:

The Effect of Ozonation on the Colloidal Stability of Particles in the Presence of Aquatic Humic Material

Principal Investigator:
Philip C. Singer

Institution:
University of North Carolina at Chapel Hill

Project Period:
2 years

Budget:
(2 years) $181,177

EPA Project Number:
R 810235

The objectives of this research are to evaluate the colloidal stability of various particulate substances in the presence of extracted humic material, and to assess the impact of ozonation on the stability of the particles. The basic goal of the research is to determine the mechanism(s) responsible for the "microflocculation" phenomenon observed when raw water is ozonated as a first step in drinking water treatment, and to identify the water quality parameters which contribute to the microflocculation phenomenon.

In the study, purified clay particles (Kaolinite and montmorillonite) and well-characterized metal oxides (alumina and silica) will be used as the particulate phases. Well-characterized humic and fulvic substances extracted from natural waters will serve as the natural aquatic humic material, and experiments will be performed in the presence and absence of calcium and ferrous iron, at constant ionic strength and in pH-buffered solutions. Colloidal stability will be assessed using particle counting and electrophoretic mobility measurements. Ozonations will be carried out on the suspensions in the presence of the aquatic humic material to determine the impact of ozone on the colloidal stability of the particles.

It is anticipated that the aquatic humic material will be adsorbed by the particulate phases and that the amount of humics adsorbed will be enhanced in the presence of calcium. It is believed that the extent of adsorption will be greatest for alumina compared to the other solid phases. It is expected that the adsorbed organic material will make the colloidal particles more stable (less susceptible to coagulation), but that upon ozonation, the organic coatings on the particles will be partially oxidized, causing them to be desorbed from the particles and thereby decreasing the stability of the particles and making them more susceptible to coagulation. In so simulating the microflocculation phenomenon, we hope to be able to better define this phenomenon and the factors contributing to it.

Title:
Molecular Mechanisms of Chemically-Induced Mutagenesis

Principal Investigator:
Gary J. Smith, Ph.D.

Institution:
University of
North Carolina at
Chapel Hill, NC

Project Period:
3 years

Budget:
(2 years) $140,104

EPA Project Number:
R 809604

The objective of this project is to gain a better understanding of how a human cell copes with chemical damage in its genetic material. There appear to be two major classes of consequences to the replication of chemically-damaged DNA. The most common perturbation of genetic integrity results from the interaction of the replicative polymerase with an isolated chemical lesion, presumably resulting in a mutational event confined to a small sequence of DNA. The alternative class of alterations of DNA sequence/structure results from the rarer occurrence of multiple sites of chemical damage within a segment of DNA; and is generally cytotoxic. The resulting perturbation of DNA replication is postulated to be associated with the induction of chromosomal mutations such as inversions, deletions, or translocations. Mutational events occurring in a single gene will be studied in the human lambda (λ) immunoglobulin gene. Clones producing a chemically-induced mutant protein will be identified electrophoretically. Culture medium from the exposed clones which contains the secreted λ protein will be isoelectrically focused and the banding pattern of the λ protein visualized by ELISA staining of a nitrocellulose blot of the focusing gel. The λ protein secreted by mutant clones will be purified, tryptic digested, the aberrant fragments identified, and the amino acid sequence of the aberrant fragments established to allow analysis of the chemically-induced change in the DNA sequence of the λ gene. Chromosomal mutational events are studied in the HGPRT locus. Clones which have concurrently lost enzymatic activity in the selectable locus, and in a linked locus, following chemical exposure will be subjected to prophase banding karyotypic analysis. The banding analysis will identify the nature of the chromosomal mutation (i.e., deletion, translocation, etc.) as well as identify the new location of translocated fragments.

The results of such studies will enhance our understanding of chemically-induced genotoxicity in two areas. First, these studies will identify the nature of the sequence alterations produced in the daughter DNA by a variety of chemicals. Second, comparison of mutation frequencies measured as isoelectric variants in the λ locus and by loss of enzymatic function in the HGPRT locus will provide insight into how much genetic damage is absorbed by the genome and not phenotypically expressed.

Title:
DNA Damage, Repair and Replication in Cell Specific In Vivo *Carcinogenesis*

Principal Investigator:
Steven M.
D'Ambrosio, Ph.D.

Institution:
Ohio State
University Research
Foundation

Project Period:
1½ years

Budget:
(1½ years) $159,477

EPA Project Number:
R 809580

The main objective of this proposal is to determine the relationship of DNA damage and repair in specific susceptible and resistant mammary cell subsets to carcinogenesis. Our approach will utilize: a) combined expertise of several interdisciplinary areas; b) rat mammary gland during different stages of development and susceptibility; c) separation of the cell types comprising the mammary gland by density gradient and elutriation techniques; d) two classes of potentially carcinogenic and noncarcinogenic alkylating agents; e) detection and quantitation of the specific DNA myoepithial; f) repair studies to follow the loss or persistence of the damage induced; and g) fixation of the DNA damage as a function of DNA replication in each cell types. The mammary gland was selected because it is one of the few adult organs with differential stages of susceptibility during development to chemically induced transformation and contains unique cell types which have the potential to transform as well as those that do not. The carcinogens used are chosen for having different potentials for inducing mammary cancer and producing different levels and types of DNA damage. The DNA damage and repair assays will be performed to correlate the particular damage induced, repair of the damage and DNA replication potential to the type of tumor arising from the different cell types of the mammary gland. All parameters employed in this study for the assessment of DNA damage and repair will be measured as functions of time, dose of carcinogen, and age of rat at time of exposure. The number of alkali-labile sites and modifications at the endo and exo-cyclic nitrogens and oxygens of DNA bases will be quantitated by monitoring the radiolabeled adducts using Sephadex and high pressure liquid chromatography (hplc) techniques. The number of alkali labile sites (phophotriesters, apurnic, apyrimidinic and breaks) as well as N-3 and N-7 alkylated purines and O^2 alkylated pyrimidine will also be quantitated using alkali and/or site specific enzymes in conjunction with sucrose sedimentation of non-radiolabeled DNA. The type of location of mammary neoplasms induced will be characterized and correlated with the level and type of DNA damage induced. Thus, the extent and nature of DNA damage, repair and replication in the susceptible and resistant cell type(s) and tumor incidence caused by several related carcinogens should help to establish: a) the relationship of DNA damage-repair to initiation; and b) DNA replication to fixation events.

Title:
Fate of DNA Damage in Human Fetal Cells

Principal Investigator:
Steven M. D'Ambrosio, PhD

Institution:
Ohio State University Research Foundation

Project Period:
3 years

Budget:
(2 years) $174,195

EPA Project Number:
R 810146

The objective of this renewal grant proposal is to further characterize at the molecular level of repair of DNA damage in human fetal cells. This will be accomplished by: relating unscheduled DNA synthesis (UDS) and scheduled DNA synthesis (SDS) directly to carcinogen binding and adducts formed; to the activity of various DNA repair enzymes (*N*-glycosylase, endonuclease and alkyltransferase) in human fetal dermal, brain and kidney cells. Human epidemiological data indicates that the nervous system and kidney exhibit high levels of cancer in the newborn. Cells from these fetal tissues also show the most significant differences in UDS. DNA containing specifically radiolabeled, M7 and O^6-alkylguanine or N3-alkyladenine will be used as specific substrates to quantitate and enzymatic activities in the cells. We also have a unique resource of human fetal tissue and culturing techniques that will allow the successful completion of this project. These studies should thus further characterize the fate of DNA damage and DNA repair in human fetal cells obtained from dermis, brain and of DNA damage and DNA repair in human fetal cells obtained from dermis, brain and kidney. The studies should also provide insights into the role that DNA damaging agents play in human transplacental carcinogenesis, teratagenesis and mutagenesis.

Title:
Development of a Sensitive Assay System for Monitoring Viral Pesticides

Principal Investigator:
George S.
Beaudreau;
Co-principal
investigator,
Margot N. Pearson

Institution:
Oregon State
University

Project Period:
2 years

Budget:
(2 years) $51,176

EPA Project Number:
R 809460

Nucleopolyhedrosis viruses (NPVs) cause a well-defined cytopathology in insects and demonstrate only a limited cross-infectivity which is normally confined to genera within the same family. Because of their restricted host range, NPVs offer an attractive alternative to chemical insecticides and therefore will probably be used in increasing amounts for pest control. One such viral insecticide, TM Biocontrol-1, has been registered by the EPA for use in controlling Douglas-fir tussock moth infestations.

It is the overall objective of this investigation to develop a specific and sensitive antibody assay system to detect viral protein. This system will provide a much needed assay for evaluating viral insecticide potency and purity and for setting standards for insecticidal preparations. It will be extremely useful for monitoring production and application of viral insecticide.

In order to develop this assay system we will: 1) prepare monoclonal antibody directed against amino acid sequences within the polyhedra protein (polyhedrin) from the nuclear polyhedrosis virus infecting the tussock moth; 2) develop an antibody assay system using these monoclonal antibodies that can both rapidly detect low levels of this virus specific protein and distinguish it from antigenically related polyhedrins of other insect viruses.

An antibody assay system using monoclonal antibody will provide a rapid and relatively simple method for detecting the presence of viral specific protein in areas that have been treated with viral insecticides. It would also be useful for monitoring the spread, if any, of the applied virus into non-host insects and other organisms. Application of this assay system should provide useful information for the development of safe management of viral insecticides. Most important, such an antibody assay system using monoclonal antibodies would provide a rapid and accurate method for monitoring viral insecticide production.

Title:
Effects of Pesticides on Baculovirus Development and Genome Stability

Principal Investigator:
William J. McCarthy

Institution:
Penn State
University

Project Period:
3 years

Budget:
(2 years) $97,033

EPA Project Number:
R 809453

The goals of this project are to: 1) evaluate the stability of baculovirus genomes to chemical pesticides by determining the effects of exposure on phenotype and genotype, and 2) identify compatible pesticide-virus combinations.

Major objectives are:

1. To establish if pesticides enhance, inhibit or have no effect on baculovirus development in vitro as determined by polyhedra (PIB) formation and extracellular non-occluded virus (NOV) titer.
2. To assess the mutagenic potential of pesticides on baculovirus genomes *in vitro*. The following classes of mutants will be screened for: a) plaque morphology mutants—those that differ from the wild type in cytopathic effects and appearance of plaques at the optimum growth temperature; b) host-range mutants—those with an increased or restricted host-cell range; and c) temperature-sensitive (ts) mutants—those that fail to replicate at higher, non-permissive temperatures while doing so at a lower, permissive one. Theoretically, ts mutants can be obtained for every gene product with an indispensable function for virus production, therefore, these mutants provide the best index for calculation of mutation frequency.

Additional objectives relevant to this project are: 1) to determine the effect of host cell genetics on baculovirus genotype and phenotype after pesticide treatment by studying one virus isolate in two different permissive cell lines; 2) to compare the mutation frequency and range of phenotypic expression with two different baculovirus isolates (a multiply-embedded isolate—MNPV) and (a singly-embedded isolate—SNPV) and; 3) to compare the compatibility of pesticide-virus combinations according to the type of baculovirus isolate (MNPV or SNPV) and cell line selected.

Title:
Validation of the Wasp, Bracon hebetor, for Testing Potential Genotoxic Chemical Agents

Principal Investigator:
Robert M. Petters

Institution:
The Pennsylvania
State University,
University Park,
PA 16802

Project Period:
2 years

Budget:
(2 years) $177,990

EPA Project Number:
R 809712

Project Objectives:

The primary objective of this study is to validate *Bracon hebetor* as an assay system for routine screening of chemical and physical agents in order to determine their potential physiological and genotoxic properties. This will be accomplished by comparing the effects of chemicals with different mutagenic modes of action on reproductive performance and mutagenic response in *Bracon hebetor*. Standardization of the *Bracon* test will allow better comparison with previous studies and future investigations. The *Bracon* test system will be evaluated for its relative effectiveness and utility through a comparison of results to those of other genotoxic assay systems.

Method of Investigation and Expected Results:

As an *in vivo* assay, *Bracon hebetor*, will permit the evaluation of chemical agents which have beem exposed to natural metabolically activating and/or inactivating systems. Since *Bracon* is an eukaryote, it will provide a genetic target similar to that of mammalian organisms in terms of its genome organization and functional properties. *Bracon* studies are ideally suited for distinguishing between general physiological insult, effects due to altered protein synthesis, and damage to DNA molecules by comparing daily egg production patterns following exposure of the female to chemical and physical agents. Due to parthenogenetic development of the eggs, treated male studies allow the detection of physiologically based sperm inactivation or genetically-determined dominant lethality. The assessment of recessive mutation induction can be accomplished for the entire genome within one generation owing to the production of haploid males from unfertilized eggs. Gross chromosome damage can be distinguished from induced recessive lethals by scoring embryonic death *in situ*.

Title:
Studies of Non-cultivatable Agents of Viral Gastroenteritis

Principal Investigator:
Robert Dolin

Institution:
University of Rochester

Project Period:
2 Years

Budget:
(2 years) $89,845

EPA Project Number:
R 809133

The overall purpose of the proposed studies is to generate information which will lead to the design, development, and evaluation of control measures for a significant public health problem, acute viral gastroenteritis, which appears to be, at least in part, waterborne. The studies are intended to represent a systematic investigation of the Snow Mountain Agent, a new agent recently described by the principal investigator. To the best of our knowledge, no other individuals or groups are studying this agent, hence the proposed studies represent a unique set of investigations in this area. Although the experiments are to be performed with the Snow Mountain Agent, it is clear that such studies may have direct applicability to other occult agents of viral gastroenteritis, such as the Norwalk or Hawaii agents. The principal investigator has accumulated significant amounts of material from volunteer studies with these other agents, and techniques which prove successful in the study of SMA could be applied directly to study of Norwalk and Hawaii agents as well.

Title:

Halogenated Aromatic Hydrocarbons: Biochemical Mechanisms of Toxicity

Principal Investigator:
Dr. Thomas A. Gasiewicz

Institution:
University of Rochester
Rochester, NY

Project Period:
2 years

Budget:
(2 years) $175,411

EPA Project Number:
R 809627

Objectives:

The overall objective of the research described is to better understand the biochemical mechanism(s) by which 2,3,7,8-tetrachloroidibenzo-*p* dioxin (TCDD) and related compounds produce toxicity in animals. More specifically, studies will determine whether altered absorption or utilization of nutrients from the gastrointestinal tract mainly contributes to weight loss and subsequent lethality in TCDD-treated animals. This will be tested by determining the effect of total parenteral nutrition (TPN) on weight gain, gross histopathology, and mortality in TCDD-treated guinea pigs. Other studies will examine if the altered disposition of some nutritional factor may contribute to hepatic damage and lethality in TCDD-treated rats. This will be performed by specific alteration in the composition of the TPN diet as well as promoting iron deficiency by the inclusion of a specific iron chelator in this diet. Other proposed studies will more clearly define and quantitate the dose and time related changes in serum and hepatic lipid alterations in TCDD-treated rats and mice. Emphasis will be placed on the various forms of cholesterol which are altered. These studies will be extended to characterize the effect of TCDD on lysosomal acid lipase which hydrolyzes cholesteryl esters, and will determine if this effect is specifically on the lipase or on general lysosomal function.

Expected Results of Research:

In view of the potential human exposure to this class of chlorinated compounds, and the observed effects of TCDD on lipid metabolism in both humans and animals, additional studies are needed to examine the biochemical alterations for these effects and the mechanism whereby TCDD produces them. In the TCDD-treated rat disturbances, the cholesteryl ester levels appear to be an early response, occurring within a few days following TCDD exposure and well before evident histological hepatic damage. The further investigation of these biochemical events may lead to an understanding into the mechanisms of TCDD-induced lipid alterations and hepatic damage. For example, it would be of particular interest to determine if an inhibition of lysosomal lipase is due to a general or specific effect upon lysosomal function. The proposed studies will examine the biochemical events responsible for TCDD-induced cholesterol alterations in rats and mice and will determine whether these alterations may be related to the action of TCDD via its cytosolic receptor.

Title:
Visual Indices of Neurotoxicity

Principal Investigator:
William H. Merigan

Institution:
University of Rochester Medical Center

Project Period:
3 years

Budget:
(2 years) $231,147

EPA Project Number:
R 809555

This research project will examine the visual system effects of three important neurotoxicants in primates. Our studies are designed to reveal the time course and nature of neuronal changes and their impact on visual capacities. A related aim is the development of sensitive clinical tests of incipient neurotoxicity for human populations. The chemicals to be studied include triethyl tin, a demyelinating agent; 2,5-hexanedione, which damages optic nerve fibers; and carbon disulfide, which causes both vascular and neuronal changes in the visual system.

Visual thresholds will be used to assess alterations in visual capacities for detecting patterned and luminance varying displays. Visual evoked responses will reveal alterations in nerve conduction in the visual pathways. Fluorescein angiography and fundus photography will be used to detect retinal vascular damage. Detailed morphological studies will be conducted on all exposed monkeys.

We expect to find characteristic patterns of visual dysfunction for each of the three agents, and determine the morphologic basis for the type of observed losses. For example, we expect that carbon disulfide will produce visual acuity losses due to damage to papillomacular bundle fibers. We are eager to determine the role of vascular lesions in this disorder. On the other hand, triethyl tin may delay visual evoked responses (by virtue of its effects on myelin) without altering pattern vision.

Title:
Pollution of Gravel Spawning Grounds Due to Fine Sediment

Principal Investigator:
Gary Parker

Institution:
St. Anthony Falls
Hydraulic Laboratory

Project Period:
2 years

Budget:
(2 years) $167,600

EPA Project Number:
R 808683

Salmon and related species of fish use the structure of natural gravelbed rivers for the purpose of spawning. Of all the factors that could adversely affect spawning success, fines accumulation ($d < 2.0$ mm) is one of the most important. In order for a new generation of fry to be produced the subpavement must not be so fine as to a) crunch the eggs in its interstices, or b) reduce groundwater flow to the point that sufficient oxygen cannot be supplied to the eggs, or metabolic wastes cannot be removed, or c) entrap the fry as they try to escape upward.

The reason for the paramount importance of fines is that of all factors deleterious to spawning likely to be exacerbated by the activities of man, it is one of the most common. Road building, forestry and agricultural activities can lead to the input of fine material into streams that exceed the natural input by one or more orders of magnitude.

A relation predicting the characteristics of retention of fines in the substrate as a function of fines input and other factors, would be of great importance since it will enable us to evaluate the impact of certain activities in stream sedimentation, and establish regulations that would preserve water quality adequate for fish survival. This problem can be approached by carefully monitored experiments that can reproduce accurately real stream situations. Here the following questions are to be addressed:

1. For given equilibrium movable-bed conditions in a stream, what is the relation between fines infeed rate and upper substrate fines content?
2. How long is required for fines content to reach equilibrium, and what is the mode of intrusion?
3. What is the effect of fines infeed on bedload transport of coarse material and on other parameters such as slope and depth?
4. How do such parameters as bed stress, fines grain size and fall velocity, and pool-and-riffle structure affect fines retention in the bed?
5. If fines input is ceased, how, and to what extent, can a stream naturally purge itself of fines?

The first step, real stream modeling, has been performed at the St. Anthony Falls Hydraulic Laboratory in a previous project funded by the EPA. The second step is expected to be approached in this project.

Title:

Luminescence Studies of the Photophysical Characteristics and Analytical Figures of Merit of Polynuclear Aromatic Compounds and Pesticides in Micellar Solutions

Principal Investigator:
L. J. Cline Love

Institution:
Seton Hall University

Project Period:
2 years

Budget:
(2 years) $157,871

EPA Project Number:
R 809474

The analytical utility of micelle-stabilized room temperature luminescence (MS-RTL) for the identification and quantitative determination of hazardous and toxic chemicals, *viz,* petroleum-based polynuclear aromatic molecules, pesticides and herbicides, and polybrominated or polychlorinated biphenyls (PBB, PCB) will be determined. The primary objective is to determine the optimized analytical figures of merit (sensitivity, useful concentration range, interferents) in order to evaluate the practical usefulness of micelle-mediated luminescence analysis techniques. These studies will answer fundamental, theoretical questions regarding the role of micelles in modifying luminiscence characteristics of molecules, and will allow development of practical, much improved analysis methods for substances which can be catastrophic to human health and well-being. These new, improved spectroscopic analysis methods will be used to screen and quantitate several types of hazardous species present in wastewaters and natural waters.

The investigative methodology can be conveniently divided into four parts. First, the physicochemical basis and experimental requirements for MS-RTL will be determined by measurement of energy/intensity spectra, quantum yields, and lifetimes for both fluorescence and phosphorescence emissions of light from the toxic molecules. Second, the chemical conditions required for observation of MS-RTL will be simplified by investigation of heavy atom effects. Third, the analytical figures of merit will be determined for a large number of hazardous species. Finally, specific analysis methods will be developed and applied to "real" environmental samples such as wastewaters and navigable waters.

These studies potentially can produce an analytical technique with enormous advantages over existing methods for the sensitive and selective analysis of environmental and forensic samples at the parts-per-billion level. This results partially from the protective micellar media used in which fewer interferents are observed and for which aqueous samples are ideal. These techniques will aid in determining compliance with water quality standards, and could have important implications for future, convenient field monitoring. The fundamental studies of MS-RTL using non-toxic heavy atoms would further simplify sample handling and disposal.

Title:

Comparative Embryotoxicity of Cadmium (Cadmium Chloride) and Mercury (Mercuric Chloride) to the Family Percidae (Class Osteichthyes, Subfamily Etheostominae, Genus Etheostoma)

Principal Investigator:
Dr. John W. Sharp

Institution:
Southeast Missouri
State University
Department of
Biology

Project Period:
2 years

Budget:
(2 years) $56,437

EPA Project Number:
R 810112

The proposed research is designed to describe the embryotoxic effects of cadmium (cadmium chloride) and mercury (mercuric chloride) to the freshwater teleosts, *Etheostoma caeruleum* and *E. spectabile,* thus demonstrating the utility of these percid fishes for evaluating potentially hazardous materials. The literature indicates that relatively few species of freshwater teleosts are used for such developmental investigations.

Initially, normal, pre-hatch stages of development will be described for each species. A description of the morphological changes and growth patterns from hatching to the juvenile stage will likewise be presented. In addition, short-term chronic biossay examinations involving adult exposure to the two metals will determine adult tolerance levels (96-hr. LC50).

Two general embryonic bioassay experiments will be run for each species and each metal. For each treatment and concentration, four replicates of 25 embryos each will be used. In the first series of experiments, 4-8 cell stage embryos will be subjected to a variety of concentrations of each metal for different lengths of time. For each concentration tested, 4-8 cell stage embryos will be exposed continuously and for 1, 2 and 5 days. Except for the continuously exposed embryos, all others will be allowed to continue development in metal-free water. The resulting data should reveal not only *concentration-dependent response differences* but also, *duration-dependent response differences.* A comparison of embryonic responses between the two species following comparable treatment to the same metal could reveal *species-dependent response differences.*

In the second series of experiments, different staged embryos (different ages), 4-8 cell, mid-blastula, mid-gastrula and beating heart embryos will be exposed chronically to each metal to describe *stage-dependent response differences* (=differential embryonic sensitivity). As above, concentration-dependent, species-dependent and toxicant-dependent response differences should be identified. For each series of experiments the following embryonic responses will be measured: mortality, anomalous development, heartbeat rate, hatching rate and total hatching success, viability of hatch and eleutheroembryonic size at time of hatching.

Experiments will be continued for 30-60 days post-hatch, with all surviving eleutheroembryos remaining exposed throughout this post-hatch period, to the media type from which they emerged. This post-hatch time period of exposure is apparently sufficient to determine the MATC (maximum acceptable toxicant concentration).

An additional experiment is designed to determine the short-term toxicity (96-h LC50) of each heavy metal on previously unexposed eleutheroembryos (yolk-sac fry) to compare the sensitivity of eleutheroembryos to the four embryonic (pre-hatch) stages. In addition, the effects of pre-exposure to these heavy metals on the sensitivity of eleutheroembryos will be fed a diet consisting of living brine shrimp nauplii, supplemented with natural food items. All media will be replaced daily.

Metal concentration in freshly prepared media, and 24-hour incubated media will be examined by atomic absorption spectrophotometry. Various EC50 values (median effective concentration) will be determined by probit analysis following correction for natural mortality. EC50 values include 96-h LC50 (mortality), SH50 (hatching success) and VH50 (viability of hatch) values. An analysis of variance will be performed on all tests. When applicable, Duncan's New Multiple Range test will be used for comparison examination. These will include concentration-dependent response differences, duration-dependent response differences and species-dependent response differences.

Title:
Heavy Metal Effects on Gene Expression in Human Cells

Principal Investigator:
Michael Karin

Institution:
University of
Southern California

Project Period:
2 years

Budget:
(2 years) $127,523

EPA Project Number:
R 810531

The objective of this project is the understanding of the molecular basis of the regulation of trace metal metabolism in human cells. The focus of the studies would be on the matallothionein (MT) system. Those low molecular weight heavy metal binding proteins play a central role in maintenance of essential trace metal (Zn^{++} and Cu^{++}) homeostasis and in protection against the harmful effect of the toxic heavy metals (Cd^{++} and Hg^{++}).

We will obtain cDNA clones of the various MT mRNA's and use them to isolate bacteriophage clones of the various genes. The structure of the mRNA's would be determined using rapid nucleotide sequencing techniques. The structure of the genes in normal human somatic cells would be compared to their structure in fibroblast lines from patients suffering from several defects in heavy metal metabolism, such as: Wilson's disease, Menkey's Kinky Hair Syndrome and Acrodermatitis Enteropathica. The cDNA's will be used as probes to investigate the regulation of the kinetics of MT mRNA production in normal and mutant fibroblast lines.

The expected results will include the structure of the various MT mRNA's. Their detailed induction in kinetics in response to heavy metal exposure in normal and mutant fibroblasts and assessment of the different components involved in regulation of MT levels, such as: transcriptional activation, mRNA degradation translational efficiency and protein degradation.

Title:
Gas-Phase Reaction Kinetics of NO_x Formation from Fuel Nitrogen in Fossil Fuel Combustion

Principal Investigator:
Prof. C. T. Bowman
Prof. R. K. Hanson

Institution:
Stanford University

Project Period:
3 years

Budget:
(2 years) $203,437

EPA Project Number:
R 810019

A combined experimental and analytical study of the gas-phase reaction kinetics of fuel nitrogen species under conditions simulating the primary zone of staged fossil fuel combustors is in progress. Two related problem areas are addressed in this program:

1. The reaction mechanism for conversion of cyano species to amine species for conditions appropriate to fossil fuel combustion.
2. The role of hydrocarbon species in the post-combustion gases on fuel nitrogen chemistry.

The experimental program is being carried out in a shock tube and in a flat flame burner facility. Various optical diagnostic and probe techniques are used to monitor the concentrations of important species during the course of reaction. Measured concentration profiles are used to infer information on the fuel nitrogen reaction mechanism and kinetic parameters.

Title:

Spectroscopic, Kinetic and Thermochemical Study of Transition Metal Adsorption at the Oxide/Aqueous Interface

Principal Investigator:
James O. Leckie

Institution:
Stanford University

Project Period:
2 years

Budget:
(2 years) $254,000

EPA Project Number:
R 809561

The overall objective of this proposed project is to experimentally verify the mechanism of transition metal adsorption at the aqueous/oxide interface in carefully chosen and characterized systems. The proposed study is divided into two major task sections: 1) spectroscopic investigation of adsorbed transition metal ions using internal reflectance spectroscopic techniques and 2) kinetic study of adsorption of transition metal ions at the oxide/aqueous interface using relaxation (electric-field jump and pressure-jump) techniques. The scope of the above proposed tasks was based on a three-year time frame for completion. Due to EPA budget restrictions, this was later amended to two years with the elimination of Task 3, the calorimetric study of surface reactions.

Due to the specialized equipment needs of Tasks 1 and 2, it is anticipated that equipment design, development, and trouble shooting will require a major portion of the first year's effort. For Task 1, internal reflectance cells will be developed. For Task 2, the pressure jump and electric field jump apparatuses will need to be built. Also for Task 2, appropriate software and hardware interfacing will be developed for transient data acquisition and equipment control. In parallel, during the first year, laboratory experiments, including adsorption, surface titration, and surface characterization, will be ongoing to determine the stoichiometry of the surface reactions.

Based on the success and progress during the first year, preliminary experiments will be run during the second year to determine the nature of the surface chemical bond of transition metal ions at the oxide surface via the spectroscopic study. Preliminary kinetic studies will also be initiated to determine the rate law and mechanism of adsorption at the oxide/aqueous interface.

When the results of the second year become available, it is anticipated that an additional year of study will be desirable (as originally planned) to fully benefit from the information, techniques, and equipment modifications that evolve through the course of the research.

Title:

Influence of Physical Structure on Adsorption of Organics by Activated Carbon

Principal Investigator:
Paul V. Roberts

Institution:
Stanford University

Project Period:
2 years

Budget:
(2 years) $181,978

EPA Project Number:
R 809601

The goal of the research is to understand the effects of the physical characteristics of the adsorbent on the rate of uptake of organic compounds from aqueous solution by activated carbon. We will develop a mathematical model of diffusion and sorption in activated carbon that accounts for the measured distribution of porosity and pore dimensions of the activated-carbon particle; verify the model experimentally utilizing a range of activated carbons with different pore-size distributions and organic adsorbates of different molecular size; and distinguish between the micro- and macropore domains with regard to the relative importance of pore diffusion and surface diffusion as mechanisms of solute transport.

The results of this research are expected to improve our understanding of the influence of the internal structure of activated carbon on adsorption phenomena relevant to the uptake of solutes from aqueous solution. A fundamental understanding of this adsorbate-adsorbent relationship will assist the design engineer in the selection of an activated carbon for a specific adsorbate of concern from the wide range of available activated carbons; further the efforts to predict *a priori* the breakthrough behavior of organic solutes in activated-carbon columns based on the results of experiments conducted in laboratory batch reactors; and add to the understanding of competitive adsorption of different sized molecules by addressing the interactions of the adsorbate with adsorbents having differing pore-size distributions.

Title:
Characterization of Aqueous Solutions for Multicomponent Adsorption and Design Calculations

Principal Investigator:
Chi Tien

Institution:
Syracuse University

Project Period:
2 years

Budget:
(2 years) $263,119

EPA Project Number:
R 809482

The principal purpose of this work is to investigate the feasibility of representing aqueous solutions containing a large and indeterminate number of soluble organic specimens as solutions containing a fewer number of pseudo species. Each pseudo species is, in fact, comprised of a number of species with similar adsorption affinity as indicated by their single species isotherm expressed in the Freundlich equations.

The study can be divided into two parts. In the first, the validity of the pseudo species concept which, in fact, represents soluble adsorbates into groups, will be examined through experimental work and computer simulation. The application of the use of the species grouping will be tested both in the calculation of equilibrium concentration as well as the dynamic behavior of column operations.

In the second part of the study, an optimization-research procedure will be established such that the adsorption affinities of the pseudo species can be estimated. The application of employing the so called tracer compound experiment in expediting the search will be explored. The establishment of procedures in carrying out the tracer measurement on a consistent basis will be first attempted followed by experimental work for its validation and confirmation.

Title:
Kinetics of Aluminum Hydrolysis

Principal Investigator:
Dr. Bill Batchelor

Institution:
Texas A&M
University

Project Period:
2 years

Budget:
(2 years) $110,957

EPA Project Number:
R 809774

The principal objective of this project is to develop a model for the kinetics of aluminum hydrolysis that is applicable to the process of coagulation in water treatment. A model will be developed to describe hydrolysis reactions in both dilute (10^{-4}M) and concentrated (1M) solutions. Continuous flow completely mixed reactor experiments will be conducted for both conditions. The experiments with dilute solutions will cover a range of hydraulic retention times (5-320 min), pH values (4-8), and two temperatures (10, 25°C). The dilute (10^{-4}M) solutions will be prepared by continuously feeding concentrated (1 M Al) aluminum sulfate solutions into the reactor. Continuous flow experiments with concentrated aluminum solutions will be conducted over a range of hydraulic retention times (0.5-16 hr), degrees of neutralization ($r = 0.5 - 2.7$) and at one temperature (25°C). During both series of experiments, reactor effluent will be sampled for mononuclear, polynuclear, and solid aluminum hydroxide species. Rates of production of these species will be calculated and used to develop the kinetic model. The ability of these species to coagulate turbidity and natural organic matter will also be determined in jar test experiments.

Successful completion of this project will result in production of kinetic models to describe hydrolysis of alumina in concentrated and dilute solutions. The model for dilute solutions is directly applicable to conditions found in water treatment plants that use alum coagulation. The models should lead to an improved understanding of the chemical processes that are important in these treatment plants. Such improved understanding can result in meeting national drinking water standards more efficiently and reliably. The kinetic model for concentrated solutions is directly applicable to the process of prepolymerization of aluminum coagulants. This process has been proposed as a way to achieve improved coagulation at drinking water treatment plants. The kinetic model should form a sound basis for evaluating this alternative treatment process.

Title:
Digested Sludge: Particle Characteristics and Dewaterability

Principal Investigator:
Desmond F. Lawler

Institution:
University of Texas at Austin

Project Period:
2 years

Budget:
(2 years) $77,408

EPA Project Number:
R 810147

The objectives are (1) to obtain data on fundamental particle characteristics (particle size distribution, density, and electrophoretic mobility) and on thickening and dewatering characteristics of raw and digested sludges through measurements on samples from operating anaerobic digesters at municipal wastewater treatment plants; (2) to make similar measurements on samples from laboratory digesters to determine the effect of control variables (detention time, temperature, strength of feed, and type of feed) on both the particle characteristics and the thickening and dewatering characteristics of digested sludge.

The approach is to conduct a two-phase study consisting of (1) field measurements on samples from municipal wastewater treatment plants and (2) semi-continuous flow laboratory experiments. In both cases, measurements of the particle size distribution, particle density, electrophoretic mobility, capillary suction time, and specific resistance will be made. In the laboratory experiments, independent variables will include the temperature, detention time, strength of feed, and type of feed; other control parameters will be held constant. It is expected that this research will yield information on how control variables affect the solid/liquid separation characteristics of digested sludge.

Title:
Chemical Frequency Distributions in Rainwater—A Physical-Chemical Model

Principal Investigator:
Dr. Howard M. Liljestrand

Institution:
University of Texas at Austin

Project Period:
2 years

Budget:
(2 years) $63,869

EPA Project Number:
R 810148

Objectives:

A characterization of the pollutant frequency distribution in rainwater will be made to better estimate the frequency of extreme (high) values. The fundamental frequency distribution will be determined from historical measurements, measurements of chemical frequency distributions at the raindrop level and, to a lesser degree, mass balance models for precipitation scavenging processes.

Methods:

The methods include computer analysis of the historical data bases for rainwater concentrations. An empirical comparison of the cumulative frequency distribution for rainwater species will be made with the best fits for various frequency distributions commonly used in air pollution analysis (normal and log-normal) as well as those used for precipitation events (Gamma, Weibull, extreme value and other distributions). In a separate study, individual raindrops will be collected and analyzed for chemical concentrations according to raindrop size.

Expected Results:

The results expected include 1) an empirical characterization of the probability density functions for rainwater concentrations, with an emphasis on the ability to predict the frequency of the highest concentrations, 2) an empirical characterization of the effect of sampling strategy on observed probability density functions, 3) an empirical characterization of the effect of precipitation scavenging on air pollutant distribution functions, 4) measurements of the chemical probability density function within the raindrop size distribution, to provide information for scavenging models and provide the chemical frequency distribution at the fundamental (smallest) sample size, and 5) a box model approximation with first and second order chemical kinetics for the interpretation of these observed probability functions.

Title:

Immunologic Analysis of Heavy Metal Toxicity

Principal Investigator:
David A. Lawrence, Ph.D.

Institution:
Albany Medical
College of Union
University
Albany, NY

Project Period:
3 years

Budget:
(2 years) $153,917

EPA Project Number:
R 809349

This project proposes to investigate the effects of heavy metals on the biochemical, biophysical, and functional characteristics of lymphocytes. Lymphocyte proliferation, immunoglobulin synthesis, and the development and differentiation of immunocompetent cells for the maintenance of antibody-mediated and cell-mediated immunity will be analyzed *in vivo* and *in vitro* with microculture techniques. *In vivo* and *in vitro* assays will be employed to deterine the cellular and subcellular heavy metal-induced alterations leading to immunopathologic conditions. Specific emphasis will be given to chronic low level exposure to heavy metals and the resultant influences on (1) autoimmune diseases, (2) bacterial infections, and (3) lymphocyte physiology. Mice will be exposed orally to various concentrations of lead, nickel, and mercury in their drinking water for 2-15 weeks. In vivo metal distribution and concentration will be quantitated by metal analysis (atomic absorption). Immunopathologic effects induced by immune complexes, autoantibodies, and/or complement deposition will be evaluated with initial emphasis on haemopoietic and renal tissues. Tissue concentration of the heavy metals will be correlated to the concentrations of immunoglobulins and/or complement components. In addition, soluble serum immune complex levels will be quantitated. Heavy metals can alter immune responsiveness, but the implications of phenomena such as lead and nickel enhancement of antibody synthesis and lead enhancement of cytophilic immunoglobulin binding have not been determined. In addition, the mechanisms involved in these biologic effects as well as lead's ability to reduce resistance to bacterial infections are unknown. Heavy metal effects will be studied at the organismal level (general health; autoimmunity; bacterial clearance; antibody production; graft rejection; complement activities), the cellular level (DNA, RNA, and protein synthesis; nucleotide levels; enzymatic activities). Evaluation of heavy metal-induced alteration of the immunoregulatory pathways will aid in the analysis of heavy metal toxicity. This study will examine the "indirect" (via the immune system) toxic effects of heavy metals and the mechanisms involved so that the magnitude and importance of heavy metal effects on the general health status of an individual can be determined more accurately.

Title:

A Rapid Bioassay for Screening Pollutants and Predicting the Impact of Chronic Exposure on Protozoan Communities

Principal Investigator:
Sharon G. Berk

Institution:
University of Virginia

Project Period:
2 years

Budget:
(2 years) $71,861

EPA Project Number:
R 809799

Objectives:

The principal objective of the research is to develop and test a rapid method for assessing sublethal effects of pollutants on aquatic microorganisms. The rapid assay will measure chemosensory responses of ciliated protozoa. Data generated from the short-term tests will be examined for their predictive value in assessing community structure changes upon low-level chronic exposure to pollutants. The major objective consists of two aspects:

1. To evaluate the use of cultures of protozoa obtained from standard culture collections or from natural populations as laboratory tools in screening and predicting effects of specific pollutants on protozoan populations.
2. To use cultures of protozoa derived from a particular aquatic environment for application of the chemotaxis chamber technique as a tool for providing information on effects of pollutants on behavior of populations in natural waters.

Method:

The chemotaxis assay procedure will be carried out with the use of modified Palleroni chemotaxis plates, which are specially designed Lucite chambers containing troughs into which microcapillaries can be placed, and wells at the ends of each trough for containing suspensions of the test organisms. After fifteen minutes, results of the tests can be determined by expelling contents of the capillary tubes and directly enumerating the protozoa by light microscopy.

Expected Results:

The proposed project should provide a rapid bioassay for screening potentially hazardous chemcials released into a freshwater or marine environment. In general, it should demonstrate the value of protozoa as laboratory bioassay tools. Specifically, the assay should determine whether sublethal concentrations of a variety of chemicals will significantly interfere with the ability of protozoa to locate nutritionally important chemical cues. Data from the rapid assay should reveal the EC_{50} for various chemicals and several species of protozoa. Results of testing protozoa grown under several culture regimes should indicate the importance of growth conditions and test media to the outcome of bioassays.

Title:
Solute Transport in Soils with Large Pores

Principal Investigator:
Peter F. Germann
Assistant Professor

Institution:
University of Virginia
Charlottesville, VA
22903

Project Period:
1 year

Budget:
(1 year) $44,964

EPA Project Number:
R 810156

The objective of this research program is to visualize solute transport in rigid artificial porous media as well as in cores of undisturbed soils, both containing large pores. Thereby, the surfaces of blocks of approximately 20 x 20 x 20 cm of both material types are sprinkled with water containing an X-ray sensitive dye. The experiments are performed on a fluoroscopic device and the infiltration process is recorded on video tape. The spatial distributions of the applied dye as response to different rates and amounts of sprinkling, the actual hydraulic properties of the matrix of the porous media and in relation to the arrangement and geometry of the large pores will provide better understanding of the nature of flow partitioning into quick flow along macropores and flow through the matrix. In addition, we want to demonstrate the initiation of water flow and solute transport along large pores.

The information gained by this program will be useful to develop more realistic concepts of infiltration in soils with large openings. The attention paid to the subject of quick flow during infiltration is increasing. Geochemists as well as designers of septic tank systems and land-applied liquid waste disposal systems are interested in improved approaches to the infiltration process.

Title:
Investigation of Pollutant Transport and Dispersion Near Isolated Terrain Obstacles

Principal Investigator:
Brian Lamb

Institution:
Washington State University

Project Period:
2 years

Budget:
(2 years) $159,010

EPA Project Number:
R 809983

Objectives:

One of the most important areas of air pollution control concerns the certification of new or significantly altered power plants and smelters by state air pollution officials under the authority of the Clean Air Act. The granting of construction and operating permits for such facilities is contingent upon the applicant's ability to show, by models and measurements, that the ground level concentrations of sulfur dioxide particulates and other criteria pollutants do not exceed national standards. This has been an especially difficult and controversial matter for plants sited in complex terrain since current models are inadequate to make confident predictions in such topographical situations. Hence, there is a need for research directed toward developing and evaluating air quality models for complex terrain. This study will provide significant support to current modeling efforts through the interpretation of field data to evaluate current theories of plume impingement on hills for certain meteorological regimes that are likely to provide significant plume concentrations on and near the hill.

Expected Results:

Results from a recent tracer field study of flow around a large isolated hill will be used to test, improve, and develop models for predicting plume behavior in areas of complex terrain. Specific areas of research will include 1) further quantitative evaluation of the tracer data with an emphasis upon development of relationships between plume behavior and meteorological observation; 2) additional comparisons of model predictions with tracer data and further development of current theoretical approaches including incorporation of drainage flow and obstacle wake modules; 3) initiation of a physical modeling study of flow around Steptoe Butte both to test fluid modeling methods and to aid in analyzing the field observations; 4) interaction between WSU and the other groups involved in complex terrain research to assure complementary efforts and efficient use of resources; 5) preliminary field measurements (flow visualization and meteorological observations) at a potential field study site to be selected in line with results of items 1-4. This work will provide a better understanding of how pollutants are dispersed around terrain obstacles. The end product will be an improved capability to predict pollutant impact from sources located in areas of complicated topography.

Title:

Influence of Exposure to Air Pollutants on Emission of Hydrocarbons and Sulfur Compounds by Vegetation

Principal Investigator:
Merrill R. Pack

Institution:
Washington State University

Project Period:
2 years

Budget:
(2 years) $197,697

EPA Project Number:
R 810153

The objectives of this research are to determine if exposure of plants to ozone, sulfur dioxide, or acid precipitation enhances or otherwise influences their emissions of hydrocarbons or sulfur compounds into the atmosphere; and if changes in the emissions are found, to determine if they are associated with development of injury symptoms or other discernible deleterious effects on the plants.

Known emitters of isoprene (oaks) and monoterpenes (pines) will be exposed separately to ozone, sulfur dioxide, and simulated acid precipitation in single-plant enclosures under controlled environmental conditions. The hydrocarbon and sulfur compound emissions will be sampled over periods of days to weeks and determined by gas chromatography. The pollutant treatment concentrations will range from somewhat above to well below the minimum levels required to cause injury symptoms to develop on the plants and will therefore be similar to the pollutant concentrations that commonly occur over extensive areas influenced by local and long-range transport of air pollutants from the urban/industrial centers of the world.

Significant effects of widely-distributed air pollutants on emissions of hydrocarbons and sulfur compounds would have important implications with respect to the atmospheric chemistry involved in the formation of organic and sulfur aerosols, ozone, carbon monoxide, hydrogen, and other important atmospheric components and might provide a means of diagnosis of subtle impacts of air pollution on vegetation.

Title:
Immunotoxicology of Polycyclic Aromatic Hydrocarbons

Principal Investigator:
Stephen H. Polmar

Institution:
Washington
University

Project Period:
3 years

Budget:
(2 years) $426,317

EPA Project Number:
R 810564

The goal of this project is to determine the effects of a class of environmental chemicals, the polycyclic aromatic hydrocarbon (PAH) on the functioning of the immune system in humans. PAH are compounds commonly found in exhaust emissions and cigarette smoke. While their role in causing cancer has been studied extensively, their potentially important effects on the function of the immune system is poorly understood. The immune system is a complex network comprised of many different types of cells (e.g. various white blood cells) and organs (bone marrow, lymphnodes, spleen) which interact to prevent or combat infections and the development of tumors. Alterations in the normal functioning of the immune system also occur in allergies and in so-called autoimmune diseases such as arthritis and certain forms of kidney disease.

This research project will study the effects of PAH particularly in low doses on the functioning of the immune system in man. A wide variety of different immune functions can be studied "in the test tube" using so-called tissue culture techniques. For example, blood cells are isolate, exposed to low doses of PAH and then studied for their ability to perform their normal function in immunity. Individuals are not exposed to the PAH being studied and these tissue culture procedures permit us to study human cells directly.

Title:

Anaphase Aberration: A Unifying Method for Development of an Aquatic Animal Gene-Tox Model

Principal Investigator:
R. M. Kocan

Institution:
University of
Washington
Seattle, WA

Project Period:
3 years

Budget:
(2 years) $241,790

EPA Project Number:
R 810057

This project will use the cytogenetic technique of anaphase aberration analysis to compare the genotoxic effects of organic compounds on cells, embryo and adult animals of the same species. This technique makes it possible to directly compare damage done to chromosomes, both *in vitro* and *in vivo*, and to relate this damage to the ultimate fate of embryos and whole animals following exposure to the same genotoxic agents (benzo(a)pyrene and MNNG).

The rainbow trout was chosen as the model species for development of this system because of the extensive background material available. Continuous cell lines, primary cell cultures, embryos and fully documented genetic stock are readily available for study.

The objectives of this study are: (1) to determine how chromosome damage produced *in vitro* compares to damage which occurs in developing embryos following exposure to similar concentrations of the same compounds; (2) to determine how an increase in chromosome damage in developing embryos ultimately affects survival, hatching, fry size and teratogenicity/tumorigenicity; and (3) whether an increase in the number of genetic loci (polyploidy) can result in greater resistance to the effects of a genotoxic agent.

The results expected from this study are: (1) the relationship which exists between chromosome damage seen *in vitro* and the ultimate fate of an organism which has been exposed to similar genotoxic agents during embryologic development will be revealed (this could be in the form of survival, morphologic alterations, tumor growth on metabolic/physiologic dysfunction); (2) the development of a field monitoring system for ichthyoplankton using chromosome squashes and anaphase aberration analysis; and (3) the significance of gene duplication as it relates to genotoxicity and the survival advantages of polyploid species.

Title:
DNA Damage and Its Expression in Mammalian Cells

Principal Investigator:
Lawrence A. Loeb;
Co-Director:
Earl P. Benditt

Institution:
University of Washington

Project Period:
3 years

Budget:
(2 years) $295,484

EPA Project Number:
R 809623

This proposal centers on the mechanisms by which damage to DNA by environmental agents results in mutations. Mutations are of concern to society, since they are causally associated with many birth defects and are central to carcinogenesis. This proposal focuses on the mutagenic consequences of unrepaired DNA damage and its expression. Two interrelated aspects of DNA damage are being explored: 1) Depurination, the excision of purine bases in DNA, occurs spontaneously and is a common intermediate in the repair of DNA damage by many environmental agents; 2) Sister chromatid exchanges appear to be a common expression of DNA damage by diverse environmental mutagens and are being extensively used as an indicator of damage.

We have recently demonstrated that depurinated sites in DNA are mutagenic during copying of DNA by purified polymerase. Our evidence indicates that misincorporation opposite depurinated sites *in vitro* are not random events, but involve the preferential incorporation of the base, deoxyadenosine. Furthermore, in one biologically relevant situation, depurination is mutagenic and also involves deoxyadenosine incorporation. To quantitate the contribution of depurinated sites to mutagenicity, we are studying the interactions of three chemical carcinogens: β-propriolactone, benzo[a]pyrene diolepoxide, and aflatoxin B_1—each with bacteriophage ΦX174 DNA—and monitoring the reversion of (amber) mutations to wild type. We predict enhanced frequency of transversions, particularly substitution of deoxyadenosine, in those cases in which mutagenesis proceeds by depurination. To study mutagenicity of depurinated sites in animal cells, we plan to use viral DNA from SV-40 in a transfection assay.

With respect to sister chromatid exchanges, our goal is to understand the biochemical mechanism of this genetic rearrangement and its relationship to DNA damage. We will examine the involvement of poly [ADP-Rib] polymerase in sister chromatid exchanges and study the relationship of these exchanges to the frequency of single-base substitutions in error-prone states. These two phenomena, depurination and sister chromatid exchanges, could be related. An understanding of both these mechanisms seems a requisite for a rational risk assessment of DNA damage by many genotoxic agents.

Title:
Simulating the Effects of Toxicants on Aquatic Microcosm Systems

Principal Investigator:
Gordon Swartzman

Institution:
University of Washington

Project Period:
2 years

Budget:
(2 years) $194,744

EPA Project Number:
R 810251

This project intercompares various model formulations in the literature relative to their ability to predict the response of aquatic microcosms to the addition of various toxicants. The microcosms under study contain 10 species of phytoplankton, *Daphnia magna*, and several species of rotifer and a large amphipod *Hyalella* sp. Six replicates were run of control experiments and six replicates apiece adding various levels of selected toxicants. These include streptomycin, N-serve, malathion and copper. Nutrient levels and the biomass levels of various biota were sampled biweekly during the life of these experiments. The microcosms are characterized by a high degree of control of the environmental conditions, a high sampling frequency and very good replicability and agreement between the replicates both for the control and treatment experiments.

In our study equations have been selected to represent the processes of primary production nutrient uptake, sinking, respiration, mortality (natural, induced by the toxicants, and predation), grazing, assimilation, excretion, and nutrient recycling. The biota have been divided into functional groups and various optional equations have been chosen for some of the processes. The options generally represent disagreements in the literature over how a given process works. Parameter values for the model were estimated both from the literature, from experiments we are conducting to estimate specific parameters and from calibration to observed behavior in the control experiments and other microcosm experiments on single species or without grazers.

Title:

The Determination of Polycyclic Aromatic Hydrocarbons by High-Performance Thin Layer Chromatography in Environmental Samples

Principal Investigator:
Prof. Colin F. Poole

Institution:
Wayne State
University
Detroit, MI

Project Period:
3 years

Budget:
(2 years) $160,538

EPA Project Number:
R 808854

The object of this project is to develop a measurement method capable of identifying polycyclic aromatic hydrocarbons (PAHs) and their heteroatom derivatives by the method of high performance thin-layer chromatography (HPTLC). The PAHs to be investigated are characteristic of energy related sources, are known or suspect human carcinogens, and are recognized by the EPA as potentially hazardous. The techniques to be developed are characterized by simplicity, ease of sample handling, high sensitivity and specificity, and have a high sample throughput. The methods when fully developed should be easy to implement and very cost effective compared to current state-of-the-art methodologies.

The intention is to use established methodologies for air and water sample collection and a combination of microbore high performance liquid chromatography and HPTLC for sample clean-up and analysis. It is expected that because of the complexity of environmental samples, HPTLC, even when combined with state-of-the art fluorescence detection techniques, may not be able to unequivocally separate, identify and quantify all PAHs of interest. The new combined instrument technique of microbore HPLC-HPTLC being developed in our laboratory will be used. Preliminary results indicate that sensitivity of the new methods is not a problem and that most of the development work will involve the elimination of matrix interferents. This will be pursued with a philosophy of maintaining the very high sample throughput of HPTLC, the development of methods which may be automated, and the development of methods involving the minimum number of operations to perform an analysis.

Title:
The Use of Wetlands to Modify Acid Mine Drainage

Principal Investigator:
Gerald E. Lang

Institution:
West Virginia
University

Project Period:
3 years

Budget:
(2 years) $173,184

EPA Project Number:
R 810082

In previous biogeochemical investigations of a freshwater wetland situated adjacent to an abandoned surface coal mine (Tub Run Bog), we have accumulated evidence to suggest that naturally occurring populations of sulfate-reducing bacteria may be removing acidity, sulfate, and dissolved iron from acid mine drainage as it percolates through the wetland peats. At a second site (Wymer Run Bog) treated, circumneutral mine drainage is being discharged into a freshwater wetland. The proposed study incorporates a series of complementary field and laboratory studies which will: 1) quantify the effect of both acid and circumneutral mine drainage inputs on wetland vegetational composition and abundance; 2) examine the effects of mine drainage on wetland surface water, subsurface water, and peat chemistry; 3) examine the hydrologic response of the watershed at Tub Run Bog to individual rain events, as compared to that of Big Run Bog, an undisturbed wetland, using concentrations of major ions and ^{18}O to separate stormflow hydrographs into groundwater and direct runoff components; 4) quantify rates of sulfate reduction and volatile sulfur gas evolution in peats collected from Tub Run Bog, Wymer Run Bog, and Big Run Bog; and 5) investigate the effects of variable sulfate and hydrogen ion loading on sulfate reduction and volatile sulfur gas flux rates in peats that have never been exposed to mine drainage. Results of these studies will permit a more accurate evaluation of the potential for using existing wetlands or creating artificial wetlands as low cost, low maintenance systems for the biological treatment of acid mine drainage.

Title:
Behavioral Effects of Low Level Pb Exposure

Principal Investigator:
Robert E. Bowman/
Nellie K. Laughlin

Institution:
University of
Wisconsin
Madison, WI

Project Period:
2 years

Budget:
(2 years) $220,442

EPA Project Number:
R 810129

The objectives are to study responses of adult female monkeys and their offspring to known chronic doses of low levels of lead, as a particularly relevant animal model of human lead toxicity. Certian common, clinical parameters (e.g., blood lead and zinc protoporphyrin levels) will be assessed, and behavioral tests will be conducted as the primary measures of potential neurotoxic damage from lead. The infant monkeys being studied include a group exposed to lead only *in utero* (5.5 mo gestation period) through dosage of the mother, a group exposed to lead only postpartum via nursing on the lead-dosed mothers from birth to weaning (6 mo), and appropriate controls. All infants were cross-fostered to achieve the desired design. The mother monkeys have been exposed to lead for a cumulative total of four years over the past six years. Lead dosage during the last pregnancy ranged from 6-10 mg/kg/day and produced blood lead levels ranging from 80-150 mg/dl. Infants at birth had the same blood lead levels as the mother (about 90 μg/dl) for lead-treated animals and 7 mg/dl for controls, and nursing infants averaged blood lead levels comparable to the lead in the mother's milk (50-100μg/dl for lead-treated animals and about 18 μg/dl for controls). The only observed clinical symptom from these treatments was a modestly reduced hematocrit at the highest lead dosages, and no effects were seen on body weight, growth, or appetite.

Both the offspring and the mothers are being tested on a variety of behavioral measures, including assessment of cognitive functions in the Wisconsin General Test Apparatus, tests of locomotor activity and reactivity, and tests of social behavior. These data are expected to determine in the offspring the relative effects of prenatal and postnatal lead exposure at fairly similar doses and durations, and the extent to which these effects are comparable to previous results of postnatal lead exposures in nursery-reared monkeys. The behavioral data on the mothers are expected to decide if prolonged, subclinical lead exposure during adulthood has neurotoxic effects and to establish a model for risks associated with long periods (i.e., decades) of low level lead exposure in the adult human. The data on the young animals are expected to indicate particularly whether toxic signs persist long after the termination of lead exposure, as an enduring and perhaps permanent consequence of organic damage. The correlation of observed toxic effects, and associated lead intakes, with such widely used clinical indicators as blood lead and zinc protophorphyrin levels, are expected to provide useful information for diagnostic or risk assessment in both developing and adult humans.

Title:

Scavenging of Aerosol Particles by Snow Crystals in the Atmosphere

Principal Investigator:
Dr. Pao-Kuan Wang

Institution:
University of
Wisconsin
Madison, WI 53706

Project Period:
2 years

Budget:
(2 years) $124,618

EPA Project Number:
R 809371

Each year a large amount of aerosol particles are emitted by human activities which form the particulate pollutants in the atmosphere. These particulate air pollutants not only cause unpleasant and unhealthy living environment for human beings but, thru the change in the atmospheric chemical equilibrium, may cause inadvertent change in climate. To understand and to solve this problem, we need to know *quantitatively* the injection and removal rates of these particulates.

The goal of this project is to determine quantitatively the removal rates of aerosol particles by falling snow crystals. Both experimental and theoretical methods will be used. The experimental study will involve a set-up which allows the natural snow crystals to fall through an aerosol chamber under precisely controlled conditions. The sizes, charges, shapes and concentrations of aerosol particles and snow crystals will be carefully determined. To prevent environmental contamination of the results, indium acetylacetonate, which is practically non-existent in the natural atmosphere, will be used as the aerosol material and will be generated by a modified La Mer generator. The amount of aerosol particles collected by the snow crystals will be measured by the quantitative neutron activation analysis. The collection efficiences are then determined from these measurements. The theoretical study involves computations using mathematical models. We will consider the capture of aerosol particles by the combined effect of Brownian diffusion, electric forces, phoretic forces, and hydrodynamic forces. Both flux and trajectory models will be used to determine the collection efficiencies. The computed collection efficiencies will be compared to the experimental results to test their validity.

The project will be expected to produce a set of realistic collection efficiences of aerosol particles by snow crystals. These results are directly usable for the assessment purpose. The theoretical model, verified by careful measurements, can be incorporated in a regional model to predict the aerosol budget.

Title:
Hydrocarbon Budget for a Polluted Marsh

Principal Investigator:
John M. Teal

Institution:
Woods Hole
Oceanographic
Institution

Project Period:
1 year

Budget:
(1 year) $31,067

EPA Project Number:
R 809825

The principal objective of this proposal is to obtain a budget for the hydrocarbons in sewage sludge that has been experimentally applied to a salt marsh over the last 11 years. We have a complete record of samples of the fertilizers added through the years for quantifying all of the hydrocarbons added to our experimental plots. We have sediment cores taken in a number of years and stored frozen from which we can obtain intermediate datum points for hydrocarbon budgets, and control plots in the same area of marsh for comparison. Secondary objectives include examination of initial products of hydrocarbon decomposition in the marsh sediments and the persistence (or lack thereof) as a result of the long-term, low-level pollution that has occurred. We also have samples from short-term experimental plots that have been experimentally treated for the past two years for additional comparison. Hydrocarbon content of the fertilizer from over the years will also be measured. This will provide us with a measure of the levels of total hydrocarbons applied and the amounts still present in the marsh sediments. This information will be valuable in providing a basis for the management of these pollutants in sewage sludge and a knowledge of the persistence of hydrocarbons in marsh sediments. This is of additional interest when compiled with information we have obtained on increased production in marshes with sludge-based fertilizer when considering the possibilities of nutrient reclamation and waste disposal of coastal towns in nearby marshes. This research will be carried out with the use of a thermal distillation-pyrolysis technique developed by Dr. J. Whelan which is a rapid and precise technique for hydrocarbon measurement in sediments of this nature.

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Title:

Predicting Adsorption Equilibria Using Calculated Activity Coefficients

Principal Investigator:
Wm. Brian Arbuckle

Institution:
University of Akron

Project Period:
2 years

Budget:
(2 years) $57,865

EPA Project Number:
R 810730-010

Summary:

Prediction of aqueous-phase activated carbon adsorption over a wide range of concentration has been unsuccessful. A new approach to predict adsorption equilibria will be attempted using activity coefficients. This study will extend a technique for calculating vapor and liquid phase activity coefficients (UNIFAC) to the adsorbed phase. Two main problems exist in doing this: representation of the adsorbent's chemical structure and calculation of the adsorbed-phase mole fractions. Five factors have been identified for evaluation, with sensitivity analysis to be performed when searching for the best absorbent structure. The adsorbent model developed will be used to predict temperature effects and adsorption isotherms for several additional solutes, including solids.

This study could provide a system to calculate the complete adsorption isotherm for nonelectrolytes on activated carbon, given a compound's chemical structure. Since the UNIFAC technique handles many compounds simultaneously, the system could ultimately predict multicomponent adsorption. This technique based on chemical functional groups could greatly aid our understanding of adsorptive phenomena, particularly the chemical interactions among the adsorbent surface, its functional groups, the solvent, and the solute.

Title:
Correcting Cascade Impactor for Blowoff and Cut Sharpness

Principal Investigator:
Gregory R. Markowski

Institution:
Atmospheric Research Group, Altadena, CA

Project Period:
1 year

Budget:
(1 year) $25,771

EPA Project Number:
R 810427-010

Summary

The objective of this research is to develop a mathematical technique to correct impactor data for stage cross sensitivity and particle blow-off. The application here is to an impactor employing duplicate stages which permit one to estimate the particle bounce-off and reentrainment. The extent of particle blow-off will also be checked by Scanning Electron Microscope (SEM) examination of the impaction stages.

Several data inversion routines (Busigin et al., Cooper et al., Crump, and Seinfeld, Simplex and Twomey) are cited by the authors. One of these methods will be adapted to the duplicate stage impactor. Criteria to be met by the inversion routine are the following: (1) Cut sharpness will be considered; (2) a functional form of the distribution is not assumed; (3) routine is runable on a micro-computer; and (4) inverted result insensitive to errors in the data. The inversion routine developed here will be checked with simulated data. It will also be applied to source data collected with a duplicate stage impactor.

Title:
Efficacy Studies of Residential Formaldehyde Control Measures

Principal Investigator:
Thaddeus J. Godish

Institution:
Ball State University

Project Period:
3 years

Budget:
(2 years) $184,198

EPA Project Number:
R 810682-010

Summary:

The principal objective of the proposal is to study and test the efficacy of specific formaldehyde control measures including source removal, source treatment, climate control and forced ventilation. All studies of treatment effectiveness will be conducted using a "conventional" wood-frame house and mobile home.

The effect of source removal and replacement with low formaldehyde-emitting substitute material will be determined in a stepwise fashion with one source removed at a time. As each source is removed, the effect on interior formaldehyde levels will be determined from continuous and dosimeter formaldehyde concentration measurements. Source treatments will focus on the use of coatings/sealants to reduce formaldehyde emissions from wood products such as particleboard and hardwood plywood paneling. The efficacy of humidity control on reducing formaldehyde levels in the test houses will be determined and information used to identify the optimum humidity level for formaldehyde control. The effectiveness of ventilation for a specific contaminant problem will be determined by how the formaldehyde is generated, the initial concentration, and the air moving capacity of the system. The efficacy of forced ventilation for formaldehyde control will also be tested for various source combinations.

Title:
Experimental Studies of the Heterogeneous Atmospheric Chemistry of Sulfur Dioxide

Principal Investigator:
John H. Seinfeld

Institution:
California Institute of Technology

Project Period:
2 years

Budget:
(2 years) $195,712

EPA Project Number:
R 810857-010

Summary:

One of the most persistent fundamental questions that has plagued the atmosperic science community over the last ten years is the extent to which heterogeneous reactions are important in atmospheric chemistry. A comprehensive experimental program investigating the heterogeneous chemistry of SO_2 in systems containing iron, manganese and carbon is proposed. The major goal of the proposed research is to further the understanding of the aerosol SO_2 to sulfate conversion link in the acidic deposition chain.

Two experimental systems will be used to study chemically reacting aerosols during SO_2 oxidation. The first is a continuous stirred tank reactor in which an aerosol's growth and chemical composition change resulting from gas-to-particle conversion can be measured. The second system is an electrodynamic balance in which the mass of a single, stably suspended, micron-sized aerosol particle may be continuously measured as the particle grows and reacts. Data obtained from both experimental systems will be subjected to rigorous mathematical modeling of the time evolution of aerosol mass and composition from which chemical conversion mechanisms will be inferred. In addition, the electrodynamic balance will be employed to address unresolved questions regarding thermodynamic and adsorptive properties of atmospheric aerosols. The use of two distinctly different, yet complementary, experimental techniques, coupled with sophisticated mathematical data treatment, constitutes a powerful approach for the study of heterogeneous atmospheric chemistry.

Title:
Optimal Application Cycle for Overland Flow Wastewater Treatment Systems

Principal Investigator:
Edward D. Schroeder

Institution:
University of California—Davis

Project Period:
16 months

Budget:
(16 months)
$119,655

EPA Project Number:
R 810530-010

Summary:

The objectives of this project are to determine the optimal wastewater application cycles for the removal of organic material and nitrogen in overland flow systems. Operating procedures currently used typically incorporate application periods of eight to twelve hours per day. These periods have been arbitrarily set on the basis of convenience, desire to maximize the draining period, and the fact that the first systems developed utilized similar periods and were successful. Because of removal mechanisms involved in overland flow treatment (sedimentation, bio-oxidation, and adsorption are of primary importance) and the temporal and spatial variation in aerobic/anoxic conditions during an application cycle, the amount of removal is probably a time-dependent function, that is it varies within the application cycle. As examples, organic removal would be expected to decrease as the dissolved oxygen concentration goes to zero in the slime layer at the soil-water interface. Ammonia nitrogen removal would slow as the soil adsorption sites become saturated and as the fraction of flow moving through the soil interface region decreases.

Optimal cycle characteristics may be different for organic and nitrogen removal. Data from preliminary studies on pilot scale systems have significantly varied over a two-hour application period. It appears that NO_3 present in the soil at the time of application is washed out or denitrified while ammonia is adsorbed to some saturation value and then washed through. Ammonia adsorption is hypothesized to be an important part of the nitrification-denitrification sequence in land treatment and thus factors such as rate of diffusion, permeability and draining between application cycles are significant design and operation variables. It is proposed to investigate these parameters using N^{15} depleted ammonium sulfate.

Title:

Experimental Studies of the Atmospheric Transformations of NO_x: Implications for Long-Range Transport

Principal Investigator:
A. M. Winer

Institution:
University of California—Riverside

Project Period:
2 years

Budget:
(2 years) $198,115

EPA Project Number:
R 810917-010

Summary

A two-year research program is proposed to investigate selected aspects of the atmospheric chemistry of a spectrum of important nitrogenous species. This research will be conducted with the spectroscopic, kinetic and environmental chamber facilities and techniques available at the Statewide Air Pollution Research Center.

Specifically, the proposal is to:

- Expand the currently EPA-funded studies of the kinetics and mechanisms of NO_3 radical reactions. In this investigation the focus will be mainly on the determination of absolute rate constants for the reaction of NO_3 with organics over the temperature range 250-400 K. The determination of these absolute rate constants is not only important to understanding the atmospheric role of NO_3, but will also enable existing relative rate data for the NO radical to be placed on an absolute basis.
- Determine the kinetics and products of the reactions of peroxyacetyl nitrate (PAN) with OH radicals. PAN is a temporary sink for NO_x and may play an important role in the long-range transport of nitrogen-containing compounds and in the NO_x budget of the troposphere. Absolute and relative rate techniques will be used to determine the rates of reactions of PAN with OH radicals over a range of temperature relevant to the troposphere.
- Investigate the formation and fates of organic nitrates from the reaction of organic peroxy radicals (RO_2) with NO under simulated atmospheric conditions. This will involve the determination of organic nitrate yields over a wide range of temperatures and pressures applicable to the troposphere, together with an investigation of the kinetics and mechanisms of their atmospheric loss processes (primarily reaction with OH radicals).

These studies will provide much-needed data relevant to the atmospheric transformations of initially-emitted oxides of nitrogen (NO_x) and to issues involving the long-range transport of nitrogenous species.

Title:
Enteric Virus Inactivation in Activated Sludge

Principal Investigator:
Richard L. Ward

Institution:
Christ Hospital
Institute of Medical
Research

Project Period:
2 years

Budget:
(2 years) $147,095

EPA Project Number:
R 810821-010

Summary:

Previous studies suggest that enteric viruses are inactivated during activated sludge treatment and that indigenous microorganisms are somehow responsible. Little more has been reported on viral inactivation during biological treatment of wastewater but the possible importance of this effect is evident in light of present reuse options for wastewater and sludge. The purpose of the study proposed here is to obtain a fundamental understanding of the effectiveness and mechanism of enteric virus inactivation during activated sludge treatment. The effectiveness of the process will be determined by the variety of enteric viruses that are affected by the process and by the irreversibility of infectivity loss associated with the process. Mechanism determinations will include studies on the physical, chemical, and biological changes in viral particles and their components that occur during treatment, the molecular and microbial agents in activated sludge responsible for these changes, and the means by which these agents cause their effects. Although biological wastewater treatment processes are not presently optimized for pathogen destruction, it is anticipated that the results of this study will provide basic information needed to make alterations leading to safe, effective, and inexpensive methods of viral inactivation during these processes.

Title:

Assessing the Effects of Chronic Stress on Freshwater Plant Communities and Soil Properties: A Data-Based Modelling Approach

Principal Investigator:
Barbara L. Bedford

Institution:
Cornell University

Project Period:
3 years

Budget:
(2 years) $214,399

EPA Project Number:
R 810150-010

Summary:

The objective of this research is to identify the key properties of wetland vegetation and hydrologic regimes that interact to determine distribution and growth of wetland plants, and to develop relatively simple field criteria by which to predict the response of herbaceous wetland vegetation to altered flooding patterns. This will be approached through a combination of: (1) synthesis of an existing data base from a Wisconsin site; (2) data acquisition from an Alaskan wetland site; (3) experimental manipulations of water levels in controlled environments; and (4) model development including evaluation using the data from the Alaskan wetland site. The approach employed in the Foret model will be used as a general paradigm for model development.

Title:

Plasmid Gene Transfer to Indigenous Organisms During Domestic Waste Treatment and Disposal

Principal Investigator:
Michael A. Gealt

Institution:
Drexel University

Project Period:
2 year

Budget:
(2 year) $116,000

EPA Project Number:
R 810049-010

Summary

The complete safety of recombinant DNA technology depends on the capability of waste treatment facilities to eliminate, not propagate, the engineered genetic material. Our preliminary studies indicate that transfer of multiple drug-resistance characteristics from plasmid containing, laboratory strains (EK2 containment level) of *Escherichia coli* to indigenous wastewater bacterial species does occur at a significant level under model treatment plant conditions. Therefore, we propose a program to determine: (1) the biological nature of the transferred gene material, i.e., whether the entire plasmid is mobilized from *E. coli* in the gene transfer; (2) the recombinational activity of the gene material when it is acquired by the indigenous wastewater bacterial host; (3) the characterization of indigenous bacterial host species; (4) the capability and frequency of mobilization of genetic material specifying characteristics other than drug-resistance; and (5) the alterations in the waste treatment procedures which would reduce transmission of plasmid material to the level of insignificance.

Title:

Remedial Treatment of Chronically High Volatile Acids in Anaerobic Digestion

Principal Investigator:
Richard E. Speece

Institution:
Drexel University

Project Period:
2 years

Budget:
(2 years) $199,300

EPA Project Number:
R 810633-010

Summary:

The basic purpose of this research is to determine if situations of retarded or "stuck" anaerobic digestion, i.e., high volatile acid levels (even at low feed rates) and slow recovery from the effect of a toxic agent, can be relieved or prevented by the use of a variety of inorganic nutrients. The experimental program envisions extensive laboratory or bench scale simulations using defined feeds. If these indicate the potential for success, experiments using digestion liquor from "stuck" digesters in the field will be conducted.

The laboratory simulations will use 4 feeds: domestic sewage sludge, cellulose, propionic acid, and acetic acid. One large batch of raw domestic sewage sludge will be concentrated by centrifugation, divided into aliquots, and frozen at -40°C. This will maintain feed consistency for six-month periods. The latter 3 feeds will include a mineral salts supplement of the usual minerals required for biological activity, i.e., Na, K, P, N, Cl, SO_4. One set of studies will be conducted at constant SRT (7.5 days and 15 days) with the loading gradually increased, starting from 2 g/l/day of organics. Three units for each feed will be used at each SRT. One will serve as a control, a second will receive an iron supplement, and the third will receive a supplement of Fe, Co, Mn, V, Zn, Mo, B, Ni, Tu, and Se. The performance of these units will be monitored in terms of gas production, gas composition, organics destroyed, pH, alkalinity, and volatile acids (total and individual). In addition, analysis for hydrogen level in the gas and Fe, Co and Ni in solution will be conducted. A similar set of studies will be set up in which the loading rate will be kept constant at 4 g/l/day of organics but the SRT will be gradually reduced from the initial value of 20 days. The same feeds, triplicate arrangement and analyses of unit performance will be conducted. It is anticipated that the units which do not receive the metals supplement will tend toward failure (high volatile acid levels) as the organic loading is increased and the SRT is decreased, before those that receive the metal supplement. If the control fails first, its contents will be anerobically transferred into six aliquots. These will be treated as follows: 1 control, 1 iron supplementation, 1 cobalt supplementation, 1 nickel supplementation, 1 titanium reducing agent, and 1 full complement of trace heavy metal supplementation plus the titanium reducing agent. These six units will not be fed any substrate after being set up. The recovery of these units from the failure condition will be followed by monitoring methane production, hydrogen level in the gas and volatile acid speciation.

In another phase of the program, parallel digesters operated at 15 day SRT and fed the 4 individual substrates at a rate of 4 gm/l/day of organics will be exposed to toxic materials (cyanide, chloroform, oxygen, formaldehyde, sulfide, low pH) at a level which should reduce methane production by approximately 90%. Substrate feeding will continue until the volatile acid concentration reaches approximately 3000 mg/l. Then the contents of each "failed" or "stuck" digester will be split anaerobically into six aliquots and subjected to the recovery test with various metal supplements as described previously.

In still a third laboratory test, 8 units will be set up at 15 day SRT and a 4 gm/l/day loading with four being fed raw domestic sewage sludge and four being fed cellulose. One unit in each set of four will then be spiked respectively with nothing (control), 3000 mg/l propionate, 3000 mg/l acetate and hydrogen at 10% in the gas. After one week with the feed and spike adjusted to maintain the above levels, the contents of each digester will be split into six parts. These six will receive the various types of heavy metal supplementation tested in the six unit recovery pattern described above. However, here daily adjustment of feed and spike will be conducted rather than complete batch operation as previously. In conjunction with these tests, methanogen biomass measurement will be attempted using methane production rate, ATP measurement, and F_{420} coenzyme assay.

If the studies above indicate that addition of trace metal can have a positive effect on anaerobic digestion, the concept will be tested on digestion liquor from field digesters and field biogas plants which are in a retarded or "stuck" condition. Digestion liquor will be collected from these units and anaerobically transferred to serum bottles containing the preferred metal supplements and reducing agents. The effect will be monitored by methane production and volatile acid speciation. Controls will be run.

Title:
A Single Gene Mutagenesis System in Drosophila melanogaster

Principal Investigator:
P. Dennis Smith

Institution:
Emory University

Project Period:
3 years

Budget:
(2 years) $122,094

EPA Project Number:
R 810390-010

Summary:

The objective of the project is the development and validation of a single gene mutagenesis system in *Drosophila* which has the capacity to provide exact molecular definition of induced mutagenic events in the presence and absence of known DNA repair deficiencies. By extending the analytical capabilities of currently available methodologies, we hope to provide a significant refinement in the state-of-the-art of genetic toxicology with this higher eukaryotic organism. We propose this system for studies of mutation induction because it has every element required of a complete mutagenesis system and features which are directly relevant to the study of mutational events in the human population.

The principal components of the system are:

1. A short-term, single generaton test is employed which can monitor mutation induction unambiguously in a large number of progeny, allowing adequate statistical sampling.
2. Mutational treatments are administered *in vivo* by feeding, injection or inhalation at various life cycle stages to a highly differentiated, sexually-reproducing animal with a genetic structure similar to humans.
3. An *in vivo* microsomal activation system has been shown to have many features in common with mammalian liver activation systems for the analysis of compounds requiring metabolic activation.

Title:

Speciation of Environmental Pollutants & Biological Materials

Principal Investigator:
J. D. Winefordner

Institution:
University of Florida

Project Period:
3 years

Budget:
(2 years) $95,000

EPA Project Number:
R 810387-010

Summary:

The proposed research is concerned with the development of: (1) a sensitive, selective, and qualitative identification of the specific chemical forms of a metal (free or bound) in a sample; and (2) the quantitative determination of the species in environmental samples.

The proposed analytical method is based upon the thermal evolution of chemical species from samples containing one or more metal constituents or certain non-metal constituents (such as B, P, Si, etc.) which can be detected by the plasma technique. The sample is injected into an atmospheric pressure inert gas microwave plasma and measurement recorded of the resulting atomic emission characteristic of the element of concern. By monitoring one or more elements as a function of evolution temperature, a selective thermal analysis is achieved whereby the species are identified by means of elemental composition and evolution temperature. The element is quantified by comparison of the resulting atomic emission signals with appropriate standards.

Preliminary results on an operating system have shown feasibility. The proposed research will involve development and optimization of the analytical instrument, improvement of sample preparation procedure, and evaluation of the technique on synthetic and real samples.

The ultimate goals of the proposed speciation method are to: (i) identify pollutants from industrial solid waste; (ii) identify toxic compounds in biological samples; (iii) evaluate toxic compounds in drinking and waste water; (iv) extend the study to evaluate ecosystem level effects of toxic materials, especially with regard to food chain concentration and toxicology; and (v) extend the analytical system for rapid screening of toxic compounds.

Title:

The Central Nervous System: Molecular and Cellular Factors Influencing Neural Development

Principal Investigator:
Paola S. Timiras

Institution:
Gordon Research Conference

Project Period:
2 months

Budget:
(2 months) $8,000

EPA Project Number:
R 810784-010

Summary:

The proposed Conference focuses on the application of gene cloning and monoclonal antibodies to neural specific markers. These neural specific markers are shown to exist as multigene families, a provocative finding in terms of neural regulation and differentiation.

In conjunction with these recent advances other aspects of neural differentiation and growth will be presented within the framework of sessions dealing with neural genetics, and neural molecular genetics, glial development, neurite outgrowth, cell recognition, synaptogenesis, cell interaction, hormones and growth factors influencing neural development. The format of the Conference will provide maximum interaction between the major speakers, no more than three per session, and the attendees who are encouraged to participate actively in the discussions and to present posters.

Title:
Chemistry of Fixed Nitrogen in Polluted Estuaries

Principal Investigator:
Steven C. Wofsy

Institution:
Harvard University

Project Period:
2 years

Budget:
(2 years) $218,500

EPA Project Number:
R 810219-010

Summary:

The proposed work will investigate nitrogen transformations in polluted tidal rivers and elucidate factors controlling production or loss of N_2O and NO in the environment, with particular attention to biological processes.

The proposed research will measure rates of primary nitrification (ammonium to nitrate) *in situ* in the Delaware and Potomac Estuaries by adding small quantities of $^{15}NH_4^+$ and measuring the rate of appearance of $^{15}NO_2^-$ and $^{15}NO_3^-$. ^{15}N analysis will be performed using a low cost emission spectrometer. Incubation procedures, similar to the one used by the investigators in earlier studies, will be used. ^{15}N will also be measured in ammonium fractions to determine the regeneration rates and assimilation rates of $^{15}NH_4^+$. In addition, evolution of N_2O and NO will be measured in the incubation flasks. NO will be measured using laser-induced fluorescence instruments, which can determine levels as low as one ppb. Outputs of these studies include: (1) measurement of nitrification rates with detailed studies of nutrient and dissolved gas concentrations in the Delaware and Potomac Estuaries; and (2) information on the origin of N_2O and NO.

A mathematical model obtained from the combination of a chemical kinetic model and a hydraulic model will be developed. Experimental data collected in the field and controlled experiments will be used for verification of the model.

The proposed experimental research will obtain new insight into the nitrogen chemistry of polluted waters. The proposed model for polluted estuaries should have direct application to problems relevant to the control of water pollution.

Title:
Fundamental Analytical Mass Spectrometric Studies Exploiting Laser Ionization

Principal Investigator:
James P. Reilly

Institution:
Indiana University

Project Period:
2 years

Budget:
(2 years) $173,269

EPA Project Number:
R 810896-010

Summary:

In recent EPA sponsored experiments the utility of combining a capillary column gas chromatograph with a laser mass spectrometer detector has been demonstrated. Work under this grant is concerned with improving the sensitivity and selectivity of this technique, through a series of apparatus changes. The goal is to attain sub-femtogram detection sensitivity for substituted aromatic compounds (mainly halogenated aromatics) and those containing sulfur and nitrogen heteroatoms. After these improvements have been completed and tested, additional molecular species (in particular larger and more thoroughly halogenated aromatic species) will be studied.

New methods of photoelectron spectroscopy and UV laser induced surface ionization mass spectrometry have recently been developed in this laboratory. The former technique, which involves irradiating a molecular beam with pulsed laser light, will be applied in spectroscopic studies of transient species that are of interest to atmospheric photochemists. The latter will be probed as a method of introducing non-volatile samples into mass spectrometers. The velocity distribution of ions generated by laser induced surface ionization will also be investigated in order to assess the importance of this distribution in determining resolution in a time of flight laser mass spectrometer.

Title:

Molecular-Beam/Mass-Spectroscopic Study of Chlorinated Hydrocarbon Flames

Principal Investigator:
Selim M. Senkan

Institution:
Illinois Institute
of Technology

Project Period:
2 years

Budget:
(2 years) $277,545

EPA Project Number:
R 810381-010

Summary:

Controlled thermal combustion of chlorinated hydrocarbons (CHCs) is an effective method for the treatment of chemical wastes bearing these compounds. However, our present day understanding of the detailed chemistry and mechanisms of CHC combustion is very limited, due, in part, to the limitations imposed by the experimental techniques utilized in the past. Therefore, we are proposing to study the atmospheric pressure, premixed, flat-flames of chlorinated hydrocarbon with the aid of a molecular-beam/mass-spectrometer (MB/MS) system, such that the formation and destruction of the stable, radical and short living species can simultaneously be observed. Probing the CHC flames at atmospheric pressure is possible because of the unusually broad reaction zones (1-2 cm.) present in these systems, which we have recently demonstrated in our laboratories.

Title:
A Study of Activated Carbon Adsorption in Vertically Stratified Beds

Principal Investigator:
Alexander P. Mathews

Institution:
Kansas State University

Project Period:
15 months

Budget:
(15 months) $35,660

EPA Project Number:
R 810432-010

Summary:

In activated carbon absorbers used in practice, the particle size varies along the length of the bed. An adsorption model and computer program are developed to describe such systems. The effect of a range of particle sizes in batch reactor studies is investigated to determine the effect on estimated surface diffusivities. Several methods of arriving at a mean diameter to describe the size distribution are evaluated.

Adsorption rate studies are conducted in batch reactors using several size fractions. Particle size analyses are made for each size fraction and the size distribution is used in adsorption models to estimate diffusivities. Fixed bed experiments are conducted using known size fractions in the bed, and the data are analyzed in the framework of a stratified bed model.

Title:
Permeation of Hazardous Waste Through Landfill Formulated of Clay

Principal Investigator:
H. Y. Fang

Institution:
Lehigh University

Project Period:
2 years

Budget:
(2 years) $260,845

EPA Project Number:
R 810922-010

Summary:

The goals of this research are to develop valid techniques to determine permeability of waste leachates through geotechnical materials, especially clay materials commonly used as permeation barriers. Techniques will be used to properly measure permeation of leachates through geotechnical materials under controlled conditions. These techniques will be used to generate data to formulate empirical permeation models. The ultimate goal is to develop theoretical permeation models which will allow prediction of permeation rates based on chemical characteristics of the leachate, chemical characteristics of the clay material, and geotechnical properties of the clay barrier material. These models will then be used to predict long-term stability of existing waste contaminant systems as well as to design future facilities.

Previous techniques for measuring the permeation of various solvents through clay material have been inadequate. Shrinkage and swelling, especially the former, may result in bulk transport of the solvent through the sample container which is not representative of the true situation. A modified triaxial cell assembly overcomes the problem previously encountered with shrinkage and swelling of the clay in a rigid sample container. The fundamental purpose of a triaxial cell is to control the state of stress on the sample. Clayey soils will be formulated from sand and various clays and consolidated in a triaxial apparatus at effective stresses representative of those in clay liners.

Clays which will be used to make the test liners are Kaolinite, Illite, and Montmorillonite. These are representative of the most common materials used for liners in waste disposal sites. The three materials will cover the majority of geotechnical barrier materials used in practice.

A total of nine geotechnical and soil chemistry tests will be utilized in the study. These are permeability, triaxial sheer and pore pressure measurement, Atterberg limit, grain size distribution, bulk, density, zeta potential, cation exchange capacity, x-ray defraction, and scanning electron microscopy.

Six chemicals in addition to "standard water" were identified for the initial tests. These are NaOH, HCl, aniline, acetic acid, acetone, and CCl_4. In addition to being representative of general classes of chemicals, all of these have been reported to be present in waste leachates. The initial studies will be at two concentrations. The maximum concentration level will be similar to that expected in most leachates. The low concentration should have minor, if any, effect on parameters in the Gouy-Chapman model.

The data generated as a result of the laboratory testing program will be analyzed using appropriate statistical procedures. Once the pore fluid-clay interactions are established, the predictions resulting from application of the Gouy-Chapman model will be compared with those determined in the laboratory.

Title:
Field Research on Radon Removal with GAC

Principal Investigator:
Jerry D. Lowry

Institution:
University of Maine at Orono

Project Period:
2 years

Budget:
(2 years) $83,861

EPA Project Number:
R 810829-010

Summary:

A two-year field and laboratory research project will demonstrate and determine the cost effectiveness of using granular activated carbon (GAC) to remove radon from private ground water supplies. A second objective is to establish a design model for future applications of the technology to municipal water treatment applications. At least three different GAC products will be used in 27 steady state adsorption/decay runs in the field. A discretized GAC bed will be utilized in the laboratory to establish the kinetics of steady state adsorption/decay as a function of steady state radon concentration in the bulk solution. These data will lead to a predictive model to determine the treatment performance characteristics for any influent radon mass loading rate.

Title:

Regional Scale Receptor Modeling in the Northeast Quadrant of the U.S.

Principal Investigator:
Glen E. Gordon

Institution:
University of Maryland

Project Period:
2 years

Budget:
(2 years) $168,835

EPA Project Number:
R 810403-010

Summary:

Receptor modeling is becoming increasingly successful for determination of the sources of particulate matter in urban air, but has been used very little on a regional scale (100s of km). Rahn has used the ratio of non-crustal Mn to non-crustal V, $(Mn/V)_{xs}$, as a tracer of Midwestern air masses into the eastern U.S. in an attempt to determine the origins of sulfate and acidity. While this is a promising approach, there are many uncertainties that must be addressed in order to place regional scale receptor modeling on a firm foundation. If that can be done, the method will be of considerable value in tracing pollutants on a scale of 100s of km. As a first step in the development of regional scale receptor modeling, the research community needs a large, very complete data set obtained in rural areas. Results from the EPA Ohio River Valley Study (ORVS) meet nearly all of the criteria except that elemental concentrations were determined by x-ray fluorescence (XRF), which is not sensitive enough to observe concentrations of a number of trace elements that may provide clear indicators of particles from certain areas or types of sources. This project will use the XRF data and trajectories for the sampling periods of the ORVS samples to identify about one-third to one-half of them for further analysis by instrumental neutron activation analysis which, combined with the XRF data, will provide results for up to 40 elements. The resulting data will be used to investigate a number of fundamental questions that must be answered before regional scale receptor modeling can be used with confidence, e.g., do particles from cities bear characteristic compositions that hold up over long distances, do coal-fired plants have clear "signals" that can be identified, are there better Midwestern tracers than $(Mn/V)_{xs}$ ratios?

Title:

Removal of Non-Volatile Chloroorganic Compounds and Their Precursors by GAC Adsorption

Principal Investigator:
Neil M. Ram

Institution:
University of Massachusetts

Project Period:
2 years

Budget:
(2 years) $108,307

EPA Project Number:
R 810492-010

Summary:

The toxic and carcinogenic risks associated with the ingestion of chlorinated water may not be attributable to trihalomethanes. The possibility exists that non-volatile chloroorganic byproducts of disinfection with chlorine may be the agents of toxicological and carcinogenic risk. This is supported by the findings that non-volatile chloroorganic products of chlorination can comprise the majority of bound halogen in chlorinated water and have been associated with a positive test for mutagenicity by the Ames test. Studies, then, need to be directed towards examining treatment methods for removing non-volatile chloroorganic compounds or their precursors from water supplies. While numerous investigations have been directed towards the removal of THMs and their precursors by granular activated carbon (GAC) adsorption, very little corresponding effort has been directed in assessing the adsorptive behavior of potentially deleterious non-volatile chloroorganic compounds and their precursors, onto GAC. The proposed research will, therefore, assess the ability of GAC to adsorb non-volatile chloroorganic compounds and their precursors from aqueous solution with or without preozonation by monitoring total, purgeable, and non-purgeable organic carbon and organic halide, as group parameters, during adsorption studies. This approach seems particularly appropriate since only a minor portion of all halogenated micropollutants is amenable to gas chromatography-mass spectrometry methods of identification. Adsorption studies using these group parameters would therefore provide important information about the removal efficiency of GAC for deleterious chlorcorganic constituents or their precursors. The research will utilize both batch equilibrium studies as well as fixed bed adsorption experiments to characterize the adsorption behavior of non-volatile compounds or their precursors onto GAC which formed upon reaction with either chlorine or chlorine dioxide.

Title:

Sequential/Competitive Reactions in Catalytic Oxidation of Model Chlorinated Hydrocarbons

Principal Investigator:
Michael P. Manning

Institution:
Massachusetts
Institute of
Technology

Project Period:
2 years

Budget:
(2 years) $196,897

EPA Project Number:
R 810397-010

Summary:

The oxidation of model chlorinated hydrocarbons has been shown to proceed over chromia catalyst in the temperature range 500-800°K. Shifts in reaction rate, yield, and selectivity have been shown to occur as a function of reactant conversion, and product or intermediate inhibitor (H_2O, CO, HCl, etc.) concentrations. Similar results have been observed previously with hydrocarbon and carbon monoxide oxidation reactions inhibited by water over chromia catalysts. Catalytic oxidation experiments performed to date have shown a small amount of formation of more chlorinated species. Alkylation of one species to form larger chlorinated hydrocarbon species may also be possible. A recycle reactor which permits measurement of the chlorocarbon oxidation kinetics in the presence of uniform concentrations of all inhibitors has been developed. This reactor will be used to measure the rate of oxidation of pairs of potentially interacting model chlorinated hydrocarbons. Inlet and outlet concentrations will be measured by gas chromatography. Kinetic rate expressions will be developed by standard regression techniques.

Title:
Anthropogenic Modification of the Silica Cycle in the Great Lakes

Principal Investigator:
Eugene F. Stoermer

Institution:
University of Michigan

Project Period:
2 years

Budget:
(2 years) $113,960

EPA Project Number:
R 810396-010

Summary:

The objective of the proposed work is to determine the long-term effects of eutrophication in a Great Lakes ecosystem. Phosphorus enrichment of the Great Lakes results in silica depletion and to modification of the food chain. Because of the unusual hydrologic budget, recent data suggests that such changes occurred much earlier than previously realized. A quantitative understanding of responses is necessary to allow best management of the Great Lakes resource.

The investigator proposes a detailed analysis of diatom population succession in terms of both species composition and abundance and a chemical analysis of the amount of biogenic silica deposited in radiometrically dated cores. Available cores from areas of rapid deposition in Lakes Erie and Ontario will be utilized. Additional field sampling will be carried out in Lake Michigan because of its more complex morphometry and loading characteristics. Also, it is the lake which has most recently undergone eutrophication and silica depletion according to available historical records.

Title:

The Impacts of Background Compounds on the Adsorption of Hazardous Organics from Concentrated Liquid and Solid Waste Leachates and Seepages

Principal Investigator:
Walter J. Weber, Jr.

Institution:
University of Michigan

Project Period:
2 years

Budget:
(2 years) $140,891

EPA Project Number:
R 810486-010

Summary:

The removal of toxic and carcinogenic compounds from landfill leachates and waste dump seepages is of paramount importance, yet relatively little is known about the behavior of such compounds in actual treatment operations. A specific and fundamental deficiency exists in our understanding of the interactions of such pollutants with naturally occurring organic material, and the impacts of such interactions on their removal in leachate treatment operations.

It has been shown that a number of priority pollutants exhibit different physical and chemical characteristics in the presence of humic and fulvic acids while the behavior of others appears to be independent of background solution. This research investigates these interactive phenomena for leachate and seepage treatment operations utilizing activated carbon adsorption processes. Adsorption is an economical and reliable method for removing many priority pollutants from single-solute solutions; this research concentrates on the more realistic situation of solutions containing complex mixtures of background organics.

The results of the study are expected to have implications and findings that extend well beyond adsorption processes. A necessary part of such a study involves a detailed evaluation of numerous pollutant/humic interactions in the hope that a better understanding of the effects of humic materials on the removal of organic pollutants in all treatment processes will evolve.

Title:
Scientific Basis for the Selection of Polymers for Water Treatment

Principal Investigator:
Mriganka M. Ghosh

Institution:
University of Missouri

Project Period:
2 years

Budget:
(2 years) $72,454

EPA Project Number:
R 809807-010

Summary:

New water quality standards and recent water pollution control requirements have created a significant increase in the use of synthetic polymers in the water industry. Use of polyelectrolytes in direct filtration may result in substantial cost savings in capital expenditures caused by the elimination of settling basins and, in some cases, flocculation basins. Further, use of polyelectrolytes as the primary coagulant may minimize the problem of sludge handling. In direct filtration using polyelectrolytes, the absence of voluminous hydrolyzing metal hydroxides means longer filter runs. Currently, most often polymers are selected on a hit-or-miss basis. There is a tremendous need for developing a scientific basis for polymer selection for the water industry. The main objective of the proposed research is to study the particle-polymer interaction from the standpoints of polymer molecular weight, charge density, flocculated particle size distribution, and polymer-mixing energies, in an effort to develop a rational method for polymer selection. Such a need is demonstrated by the recent formation of a polyelectrolytes standards commitiee by the American Water Works Association.

Title:

Survival, Growth and Genetic Recombination of New Genomes in Soil and Other Natural Ecosystems

Principal Investigator:
Guenther Stotzky

Institution:
New York University

Project Period:
2 years

Budget:
(1 year) $66,277

EPA Project Number:
R 809067-010

Summary:

The survival, establishment, and growth of bacteria in natural habitats are dependent partly on their genetic constitution and, perhaps to an even greater extent, on the chemical and physical characteristics of the habitats (e.g., organic and inorganic nutrients, moisture, temperature, pressure, gaseous composition, radiation, pH, E_h, surfaces, toxicants). The impact of these physicochemical factors is greater on the establishment of introduced microbes than it is on indigenous populations. As the relative importance of each factor and of their interactions differs with the type of habitat, the impact cannot always be predicted without experimentation. Furthermore, these factors also influence the survival of "naked" and "encapsulated" DNA, i.e., used in transformation and transduction in natural habitats, even in the absence of appropriate hosts, as well as the ability of bacteria capable of genetic recombination to transfer and accept both chromosomal and plasmid DNA.

Title:
Atmospheric Reactions and Atmospheric Mutagenic Changes in Emissions from Common Fuels—An Outdoor Chamber Study

Principal Investigator:
Richard Kamens

Institution:
University of
North Carolina—
Chapel Hill

Project Period:
2 years

Budget:
(2 years) $149,991

EPA Project Number:
R 810773-010

Summary:

The purpose of this research is to study the chemical transformations and associated bioaction that particulate emissions from common fuels undergo once they are emitted into the atmosphere. Many researchers have demonstrated that the organic particle extracts taken directly from combustion sources have a measurable mutagenicity as determined from Ames mutagenic screening procedures. Little is known, however, about the transformations of these emissions once they have left the stack and age and interact with other pollutants in the atmosphere. There is some preliminary and indirect evidence to suggest that changes do occur. Very recent work conducted at the University of North Carolina in outdoor chambers has shown that the mutagenic activity of dilute soot particles can be enhanced by a factor of 2-5 after reacting wood smoke in the dark with 0.2-0.3 ppm mixtures of O_3 and NO_2. One may speculate that similar types of atmospherically induced mutagenic changes occur with emissions from other fuels such as diesel, peat and coal, but the magnitude of these changes is unknown. Hence there is a strong need to evaluate the atmospheric mutagenicity of different sources under a range of controlled, *realistic* outdoor conditions and factor this into urban air mutagenicity estimates. There is a need to identify the chemical species or groups of compounds which potentially give rise to these mutagenicity changes. This will lead to a better understanding of atmospheric transformation processes for the different emission types. Dilute combustion products will be added to two existing 25 m^3 outdoor environmental chambers from a diesel automobile and a commonly used residential wood stove, which will be used to burn peat, coal, and wood. These chamber systems will then be exposed to sub-ppm concentrations of NO_2 and O_3, both in the light and in the dark, and to photochemically active smog systems in natural sunlight. Particle extract samples will be fractionated so that 10-15 selected PAHs, nitroaromatics, quinones and phenolic compounds can be traced during an experiment. Ames type mutagenicity tests on initial and final extracts will also be conducted.

Title:

Environmental Carcinogen Perturbation of Differentiation and Induction of Carcinogenesis: Human Epithelial Cells

Principal Investigator:
George E. Milo

Institution:
Ohio State University

Project Period:
3 years

Budget:
(2 years) $159,877

EPA Project Number:
R 810407-010

Summary:

The objective of this proposal is to examine the mechanism of how airborne xenobiotic carcinogens of interest to EPA interact with human epithelial cells *in vitro*. We have selected the human lung type II cell as a target. The program for expression of a carcinogenic insult in lung type II cells *in vitro* has already been completed. What we need to understand is the relationship between the metabolic activation of specific airborne carcinogens, carcinogen-DNA adduct formation, expression of the carcinogenic insult and perturbation of the differentiated process of these target cells. Correlative support studies also will be undertaken to examine this program with the program of carcinogenesis in isogenic lung fibroblast.

Carcinogen-DNA adduct studies completed here will also be undertaken in keratinocytes to evaluate how human "skin" responds to the carcinogenic insult at the DNA level of response.

Lastly, the parameters defined for the *in vitro* transformations will be evaluated with tumor tissue from human patients to validate the indices presented here for measuring achorage independent growth, allular invasiveness and neoplasia.

Title:

Sampling and Characterization of Organic Aerosols

Principal Investigator:
James J. Huntzicker

Institution:
Oregon Graduate Center for Study and Research

Project Period:
2 years

Budget:
(2 years) $177,592

EPA Project Number:
R 810356-010

Summary:

The sampling of organic aerosol is complicated by the vapor-particulate equilibria which exists for organic compounds present in aerosols. The presence of a compound in both phases can lead to sampling artifacts which are poorly understood at the present time. Such artifacts include volatilization losses during sampling and adsorption of organic vapors onto the filler material and deposited aerosol. The proposed research will examine these phenomena in detail. Measurements of both volatilization loss and adsorption gain will be made as a function of filter face velocity. Specific compounds responsible for these artifacts will be identified by solvent-extraction GC/MS analysis. The relationship of the artifacts to the organic vapor phase composition will also be explored. From these experiments an optimum sampling procedure for organic aerosol will be devised and will be used for measuring the concentration of organic material in atmospheric aerosols.

Title:

The Contribution of Residential Wood Burning to Urban Air Pollution

Principal Investigator:
M. A. K. Khalil

Institution:
Oregon Graduate Center for Study and Research

Project Period:
2 years

Budget:
(2 years) $182,428

EPA Project Number:
R 810091-010

Summary:

Because of the rising costs of traditional fuels, many people have begun to use wood to provide all or part of their residential space heating needs. This growing dependence on wood has increased the particulate air pollution burden on many American cities. The goal of the proposed research is to develop and evaluate an experimental methodology to determine accurately the contribution of residential wood burning to particulate urban air pollution. Detailed studies of important fine particle sources (wood burning, distillate and residual oil combustion, natural gas combustion, and vehicular exhaust) will be made. Parameters to be measured include the chemical composition of the aerosol, the mass distributions of the chemical components (in particular, organic and elemental carbon), and the ratio of methyl chloride (CH_3Cl) to fine aerosol mass. CH_3Cl is a recently recognized vapor phase tracer for wood smoke. Simultaneous receptor site measurements will include the chemical composition of the fine aerosol, mass distribution of the important chemical components, and the concentration of CH_3Cl. The contribution of wood burning will be deduced in three ways: from the ambient measurement of CH_3Cl and the ratio (CH_3Cl/fine aerosol) in wood smoke; from chemical element balance calculations using the ambient chemical data and the source compositions for wood smoke and other important sources of fine aerosol; and by the size distribution balance approach. This last method is a linear least squares reconstruction of the ambient size mass distribution for organic or elemental carbon (the principal chemical species in wood smoke) from the mass distributions from the various sources.

Title:
Potential for Biotransformation of Toxic Chemicals Defined by a Molecular Probe

Principal Investigator:
Ramon J. Seidler

Institution:
Oregon State University

Project Period:
2 years

Budget:
(2 years) $126,550

EPA Project Number:
R 810645-010

Summary:

The purpose of this study is to develop and test a new method that can be used to predict the fate of specific synthetic chemicals which enter the environment. This will be accomplished by cloning genes which code for a specific structural activity group of xenobiotics. The clone genes will be used as a "reagent" or probe for directly enumerating bacteria which possess the synthetic chemical metabolizing enzymes. Within 2-3 days the size of the microbial population which is capable of metabolizing the chemical(s) will be known. The assay will be standardized by using conventional techniques for detecting such bacteria.

The technique is widely applicable for studying the fate of any group of synthetic chemicals which may enter the environment. Results of the assay can be used by the regulatory agency for predicting the relative persistence of toxic synthetic chemicals in various locations and environments.

Title:
Studies of Non-Cultivatable Agents of Viral Gastroenteritis

Principal Investigator:
Raphael Dolin

Institution:
University of Rochester Medical Center

Project Period:
2 years

Budget:
(2 years) $50,547

EPA Project Number:
R 810612-010

Summary:

The overall purpose of the proposed studies is to generate information which will lead to the design, development and evaluation of control measures for a significant public health problem, acute viral gastroenteritis, which appears to be, at least in part, waterborne. The studies are intended to represent a systematic investigation of the Snow Mountain Agent, a new agent recently described by the principal investigator. To the best of our knowledge, no other individuals or groups are studying this agent, hence the proposed studies represent a unique set of investigations in this area. Although the experiments are to be performed with the Snow Mountain Agent, it is clear that such studies may have direct applicability to other occult agents of viral gastroenteritis, such as the Norwalk or Hawaii agent. The principal investigator has accumulated significant amounts of material from volunteer studies with these other agents, and techniques which prove successful in the study of SMA could be applied directly to study of Norwalk and Hawaii agents as well.

Title:

Using the Natural Detoxification Capacities of Organisms to Assess the Environmental Impact of Contaminants

Principal Investigator:
David Brown

Institution:
Southern California
Coastal Water
Research Project
Authority

Project Period:
3 years

Budget:
(1 year) $100,000

EPA Project Number:
R 810248-010

Summary:

The loading or binding capacity of detoxification systems for trace metals or xenobiotic hydrocarbons is unknown for marine organisms. As a result, the degrees of saturation of these systems in marine organisms from the contaminated areas remain unknown. The objectives of this proposal are: (1) to develop a contaminant-specific *in situ* bioassay capable of assessing the impact of trace metals and xenobiotic hydrocarbons on marine organisms; (2) this *in situ* assay should be capable of assessing both the acute (short-term) and chronic (long-term) impact of contaminants on the organisms; (3) this project will refine current methods for assessing the partitioning of trace metals between sites of detoxification and sites of toxic action and develop parallel procedures for determining partitioning of xenobiotic hydrocarbons; and (4) the loading capacity of the detoxification systems for both trace metals and hydrocarbons will be defined.

Once loading capacity is determined, we should be able to predict how much more contaminant can be released into these environments before detoxification capacities of certain species will be exceeded, with resultant spill-over of contaminants into sites of toxic action and the resultant detrimental effect on the organism.

Title:

Flow Cytometry of Sperm as a Dosimeter of Toxic Chemical Health Hazards

Principal Investigator:
Donald P. Evenson

Institution:
South Dakota
State University

Project Period:
2 years

Budget:
(2 years) $119,337

EPA Project Number:
R 810986-010

Summary:

This project is intended to develop new flow cytometry techniques to measure minimal chromosomal damage to sperm and lymphocytes of individuals following either a single exposure or multiple low-level exposures to hazardous chemical agents. Biological markers of chemical exposure are presently inadequate for monitoring this type of exposure, but the effects on reproduction and chromosomes are believed to provide the most sensitive test (Science 215:643, 1982). Our present proposal utilizes newly developed flow cytometry techniques to assess the sensitive cell types. With these techniques, it is possible to detect and quantitate statistically, rare as well as early, adverse changes in sperm chromatin (Evenson *et al,* Science 240:1131, 1980) as well as alterations of DNA content and chromosome structure in lymphocytes in response to irradiation (Evenson *et al,* in preparation) or to mutagenic chemicals (Otto *et al,* 1981).

We intend to modify and further develop our present methodology to increase the sensitivity of flow cytometry to changes in sperm morphology and chromatin structure, comparing the measurements obtained in animals exposed to a variety of potentially toxic chemicals with observations and measurements of sperm by light and electron microscopy.

All modifications in the flow cytometry technique will be optimized by means of careful dose-response analysis. The features to be measured will include: (1) number and type of chemically-induced shape abnormalities of sperm; (2) chromatin thermal denaturation profile of sperm and lymphocytes, devised by us; (3) accessibility to various fluorescent probes of sperm and lymphocyte chromatin structure; and (4) alterations of lymphocyte chromatin and DNA content.

Title:
Long-Term Measurements of Trace Chemicals in the Troposphere

Principal Investigator:
H. B. Singh

Institution:
SRI International

Project Period:
2 years

Budget:
(2 years) $130,443

EPA Project Number:
R 810573-010

Summary:

The atmospheric burdens, growth and fates of the halogenated and many nonhalogenated pollutants proposed here for investigation are poorly understood. Significant quantities of these chemicals are released into the environment; the sources of the emissions are primarily man-made. In predicting the environmental effects of pollutants, it is also essential to study the natural atmosphere and its interaction with man-made chemicals. The possibility of destruction of stratospheric ozone by halogenated compounds and the toxicity and carcinogenicity of stable pollutants make continued research urgent. In addition, a number of these globally distributed chemicals absorb infrared radiation, which could cause a warming of the earth's atmosphere. On the positive side, many halocarbons (such as methyl chloroform) are convenient chemical tracers for studying the chemistry of the natural atmosphere.

During the proposed three-year study, biweekly measurements of some 33 selected trace constituents will be made, which will include most of the important trace species of interest, man-made as well as natural. These measurements will be made at a clean coastal site in California. Similar measurements have been made since 1979. Sufficient data will be collected to establish seasonal and long-term trends in the abundance of tropospheric constituents.

Title:
Iron Treatment by Sodium Silicate and Chlorine

Principal Investigator:
Robert B. Robinson

Institution:
University of Tennessee

Project Period:
2 years

Budget:
(2 years) $95,681

EPA Project Number:
R 810731-010

Summary:

Effective treatment of iron in potable water usually involves expensive water plants comprised of aeration, detention, filtration, chemical addition, and repumping. Another treatment technique has been pioneered in Ontario, Canada, which relies only on the addition of sodium silicate and chlorine; the small capital cost of this treatment is attractive to small towns, but this treatment has found wide use only in Ontario. The lack of wider use is perhaps due to a poor understanding of the fundamentals behind the treatment, a fear of easy solutions, and a need for a better dosage formula. This research aims to develop an understanding of the fundamentals behind this treatment method.

Title:

Continuous Determination of Ambient Hydrogen Peroxide: Airborne Measurements of H_2O_2,
HCHO and S(IV) in Cloudwater and Gas Phase

Principal Investigator:
Purenendu K. Dasgupta

Institution:
Texas Tech University

Project Period:
2 years

Budget:
(2 years) $90,103

EPA Project Number:
R 810894-010

Summary:

A continuous monitor for the measurement of gas phase H_2O_2 with a limit of detection of ≤ 0.1 ppbv H_2O_2 (g) will be developed. A continuous flow system (microflow conditions) with a fluorescence detector will be coupled with an existing diffusion scrubber. The diffusion scrubber is able to provide an air/water transfer coefficient some 500-fold better than that attained by a midget bubbler. The analytical reaction will involve the oxidation of *p*-hydroxyphenylacetic acid with H_2O_2 in the presence of peroxidase to produce a fluorescent dimer. This method will also be adapted for measurement of H_2O_2 in cloudwater. Additionally, the formation and destruction of H_2O_2 from and by O_3 will be studied as a function of $[O_3]$, pH, electrolyte concentration, presence of transition metal ions, Cl^-, unsaturated hydrocarbons and S(IV). Techniques will be developed to distinguish between HCHO bound and free S(IV) and between S(IV)-bound and free HCHO. Measurements of HCHO, S(IV) and H_2O_2 in cloudwater and the surrounding gas phase will be carried out with these methods. The ammonium salt of hydroxymethanesulfonic acid will be synthesized and its volatility measured.

Title:

Habitat Selection and Population Dynamics of Dominant Fishes in Estuarine Nurseries: A Framework for Evaluating the Role of Shallow Water Habitats

Principal Investigator:
Michael P. Weinstein

Institution:
Virginia Commonwealth University

Project Period:
2 years

Budget:
(1 year) $45,931

EPA Project Number:
R 810334-010

Summary:

The objectives of this proposal are to determine the optimum shallow water habitats for young spot and other fish species in the lower Chesapeake Bay, and to ascertain how open the populations are, once they have been recruited into the nurseries. Growth and size-at-age will be used as an index of habitat suitability. Marsh and seagrass meadow nurseries will be assessed in terms of relative habitat value.

Mark-recapture techniques will be used to determine habitat preferences, residence time and population turnover rate for spot, an ubiquitous and key species of estuarine studies.

The study site is the York River in Virginia, a major tributary of the Chesapeake Bay. It is relatively uninfluenced by human impact and offers the habitats of interest: *viz.*, extensive marshes and tidal creeks, freshwater to polyhaline zones and large seagrass meadows.

Title:

Use of Direct Freeze Conditioning Techniques to Improve Biological Sludge Dewatering Characteristics

Principal Investigator:
William R. Knocke

Institution:
Virginia Polytechnic
Institute and
State University

Project Period:
1 year and 5 months

Budget:
(1 year and 5 months)
$71,743

EPA Project Number:
R 810893-010

Summary:

The research program will examine use of immiscible gases as a potential cost-competitive alternative for improved sludge dewatering. Direct freeze-thaw conditioning techniques using hydrate forming gases (propane, Freon) will be compared from an economic viewpoint to the use of conventional (chemical) conditioning techniques. Specific objectives of the research are to: (a) develop a prototype continuous flow direct freeze reactor and optimize sludge conditioning using immiscible gases; (b) examine the impact of freeze-thaw conditioning on such sludge parameters as particle size, density, and internal water content and ultimately on the rate and extent of sludge dewatering; (c) examine interaction of refrigerant gas and the sludge to determine potential for formation of gas-water complexes; and (d) compare economics to commercially available chemical conditioning methods (lime, iron, polymers).

Title:

Evaluation of Building-Wake Dispersion Algorithms and Development of a Realistic Wake Model

Principal Investigator:
Brian Lamb

Institution:
Washington State University

Project Period:
1 year

Budget:
(1 year) $32,437

EPA Project Number:
R 810450-010

Summary:

The data from an extensive tracer investigation of pollutant transport and dispersion near an isolated building will be used in a quantitative evaluation of currently available gaussian dispersion models including the Industrial Source Complex (ISC) model and the Point, Area, and Line Source (PAL) model. The results of this analysis will be used to direct improvements in the treatment of dispersion rates and plume heights in these models.

Two recent theoretical methods describing flow around obstacles will be used as the basis for developing a more realistic wake dispersion model. One approach involves the application of potential flow theory to flow over a building with turbulent eddy diffusion incorporated in the streamline model. This model can be used to predict concentrations in the recirculation cavity and in the wake downwind of the building. The second approach employs superposition of momentum wake and vortex wake theories to predict scalar dispersion in the far wake of the building. Computer algorithms of both approaches will be developed and the models will be tested by sensitivity analyses and by evaluation against the tracer data. The results of this research will be combined to provide the best overall approach for modeling flow around buildings.

Title:
Aerosol Extinction Measurement

Principal Investigator:
Alan Waggoner/
Ray Weiss

Institution:
University of
Washington

Project Period:
1 year

Budget:
(1 year) $62,976

EPA Project Number:
R 810837-010

Summary:

Measurement and modeling of visual effects of pollutants is required under the Clean Air Act Amendments of 1977. Almost all effects are proportional to an optical parameter, the extinction coefficient, equal to the sum of scattering and absorption by particles and gases. In most cases, particle extinction determines visual air quality and there are standard techniques for measurement of scattering and absorption by particles. There appears to be some uncertainty in the scientific literature as to the accuracy of the integrating nephelometer used to measure scattering and the integrating plate used to measure absorption. The accuracy of these two extinction measurement techniques will be tested using laboratory aerosols and measurement of extinction by light attenuation over a 24-meter length optical cell. The cell sensitivity for measurement of extinction is of order 2×10^{-5} m^{-1}, about 1% of the extinction level of typical aerosol in this study. The cell has been tested using scattering aerosol by comparison to an integrating nephelometer. Measured (R^2) correlation of scattering and extinction, when the concentration of laboratory aerosol is changed by dilution, is greater than 0.9998. Absorbing aerosol will be generated from dye and from graphitic carbon to calibrate the integrating plate method and compare various filter media against Nuclepore. Internally mixed aerosol, made from a mixture of absorbing and nonabsorbing materials, will be used to model ambient particles in the tests of methods to determine absorption. Extinction and scattering by spherical and non-spherical particles will be compared to test for effects of particle shape. The results of this program will define the confidence with which extinction can be measured in the ambient atmosphere and modeled from calculated aerosol concentration.

Title:
Membrane-Covered Rotating Disk Electrode

Principal Investigator:
R. B. Smart

Institution:
West Virginia University

Project Period:
3 years

Budget:
(2 years) $61,107

EPA Project Number:
R 810540-010

Summary:

The purpose of this project is the development and application of a membrane-covered rotating disk electrode for measurement of trace metals in the environment. Artifacts due to the adsorption of organics on mercury electrodes have been a major hindrance to obtaining reliable data from natural waters by voltammetric techniques. By covering the electrode with a dialysis membrane, larger organic molecules are excluded from the electrode surface.

The membrane-covered electrode will initially be examined in the presence of compounds (such as alkaline phosphatase, Triton X, and gelatin) previously shown to adsorb at mercury surfaces. The complexing capacities and conditional stability constants of a well defined soil-derived fulvic acid, with Cu and Cd, will then be determined using the membrane-covered electrode. The results will be directly comparable to previously published data generated by other measurement techniques.

Title:

Historical Records of Pollutants in Recent Aquatic Sediments

Principal Investigator:
Erik R. Christensen

Institution:
University of Wisconsin

Project Period:
2 years

Budget:
(1 year) $57,382

EPA Project Number:
R 810419-010

Summary:

The proposed project is concerned with records of particle-associated substances (Pb, Cd, Zn, PCBs) in recent aquatic sediments. Mixing of the upper sediment layers by deposit feeders tends to smear out the input function of sedimenting substances. Similarly, compaction of sediments compresses the input record in the lower layers. The sediments thus act as a low-pass filter, allowing only slow variations in the input function to be reflected in the sedimentary record while rapid variations are attenuated.

The goal of the project is to develop mathematical models to describe these effects. The advection-diffusion equation will be used to predict the sedimentary profile based on a known input history and known sedimentation processes. Mixing and compaction will be determined by fitting the steady-state model to existing profile data. The inverse problem, to determine the input history based on a known sedimentary record and known sedimentation parameters, will be approached by deconvolution using Laplace or Fourier Transforms.

The results of the calculations will be compared with data from the literature and with measurements on sediment cores collected from Lake Michigan and other aquatic systems, possibly the southern California coast or the Hudson River Estuary. Pb-210 and Th-228 will be used as sedimentation tracers, and analysis for Pb, Cd, Zn and PCBs will be used to test the model.

Title:

Prediction of the Minimum Bed Expansion Requirement During Filter Backwashing Operations

Principal Investigator:
Jerry Y. C. Huang

Institution:
University of Wisconsin, Milwaukee

Project Period:
2 years

Budget:
(2 years) $137,150

EPA Project Number:
R 810915-010

Summary:

The current knowledge of filter backwashing, regarding the factors affecting the solids dislodgement and the required bed expansion for an effective bed cleaning, is seriously inadequate. This research attempts to establish the relevance of the properties of filtering flocs on the solids dislodgement during filter backwashing. The specific objectives are to: (1) evaluate the difference in solids dislodgement from solids flocs of varying properties; (2) correlate the solids dislodgement with the extent of bed expansion; and (3) determine the minimum bed expansion requirement for an effective bed cleaning for solids flocs of varying properties.

This research plan consists of three tasks: (1) filtering operation with various types of influent impurities and with various types of coagulant/flocculant in various ranges of dosage; (2) characterization of filtering flocs in terms of their strength and particle size distribution; and (3) backwashing operation with varying extents of bed expansion and establishment of the correlations between the bed expansion requirements and floc properties.

The research will be conducted in three phases: (1) design and construction of filtering and backwashing systems and concentric rotating cylinder apparatus; (2) filtering and backwashing of the filters applied with synthetic solids flocs, characterizing these solids flocs, and establishing the correlation between the property of the flocs and the bed expansion requirement; and (3) similar operations to those in (2), but with natural waters, for verifying the results obtained with synthetic solids flocs and assessing the difference.

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